

Elucidating the source–sink relationships of zinc biofortification in wheat grains: a review

Article

Published Version

Creative Commons: Attribution 4.0 (CC-BY)

Open Access

Xia, H., Wang, L., Qiao, Y., Kong, W., Xue, Y., Wang, Z., Kong, L., Xue, Y. and Sizmur, T. (2020) Elucidating the source–sink relationships of zinc biofortification in wheat grains: a review. *Food and Energy Security*, 9 (4). e243. ISSN 2048-3694 doi: <https://doi.org/10.1002/fes3.243> Available at <https://centaur.reading.ac.uk/92435/>

It is advisable to refer to the publisher's version if you intend to cite from the work. See [Guidance on citing](#).

To link to this article DOI: <http://dx.doi.org/10.1002/fes3.243>

Publisher: Wiley-Blackwell

All outputs in CentAUR are protected by Intellectual Property Rights law, including copyright law. Copyright and IPR is retained by the creators or other copyright holders. Terms and conditions for use of this material are defined in the [End User Agreement](#).

www.reading.ac.uk/centaur

CentAUR

Central Archive at the University of Reading

Reading's research outputs online

REVIEW



WILEY

Elucidating the source–sink relationships of zinc biofortification in wheat grains: A review

Haiyong Xia^{1,2}  | Lan Wang^{1,2} | Yuetong Qiao^{1,2} | Weilin Kong¹ | Yanhui Xue¹ | Zongshuai Wang¹ | Lingan Kong¹ | Yanfang Xue³ | Tom Sizmur⁴

¹Crop Research Institute, Shandong Provincial Key Laboratory of Crop Genetic Improvement, Ecology and Physiology, Shandong Academy of Agricultural Sciences, Jinan, China

²College of Life Sciences, Shandong Normal University, Jinan, China

³Maize Research Institute, National Engineering Laboratory for Wheat and Maize, Shandong Academy of Agricultural Sciences, Jinan, China

⁴Department of Geography and Environmental Science, University of Reading, Reading, UK

Correspondence

Haiyong Xia, Crop Research Institute, Shandong Provincial Key Laboratory of Crop Genetic Improvement, Ecology and Physiology, Shandong Academy of Agricultural Sciences, Jinan 250100, China.
Email: haiyongxia@cau.edu.cn

Funding information

Shandong Provincial Key Research and Development Program of China, Grant/Award Number: 2018GNC111012; State Key Laboratory of Crop Biology of China, Grant/Award Number: 2016KF05; National Key Research and Development Program of China, Grant/Award Number: 2016YFD0300202, 2017YFD0301005 and 2018YFD0200603; National Natural Science Foundation of China, Grant/Award Number: 31702001; Shandong Provincial Innovation Project for Agricultural Key Technology Application: Research and Demonstration of Annual Planting Patterns and Key Technologies for Green Improvement of Grain, Industrial and Forage Crops Production; Innovation Project of Shandong Academy of Agricultural Sciences of China, Grant/Award Number: CXGC2016B04

Abstract

Zinc (Zn) concentration in wheat grains is generally low, with an average value of around 28–30 mg/kg. Therefore, increasing wheat grain Zn concentration for better human health is the focus of HarvestPlus global initiatives. Source–sink interactions have been intensively studied for decades to enhance crop yield potential, but less on grain nutritional quality. This review applies concepts of source, sink, and their interactions to the study of wheat grain Zn nutrition and biofortification. Increasing Zn sources to wheat (via soil and foliar application) could directly enlarge available Zn in vegetative tissues and grain Zn sink. Rational nitrogen (N) supply increases grain Zn accumulation (N–Zn synergism), but phosphorus (P) input generally decreases (P–Zn antagonism), and the potassium (K) effect is unclear. Conventional and genetic breeding have potential to stimulate Zn flow from source to sink (uptake from soil, root-to-shoot translocation, and remobilization). However, a rational manipulation to establish a well-coordinated source–sink relationship is required to finally realize the grain Zn target (40–50 mg/kg) and increase on-farm crop yield. Future studies should focus more on fertilization modes adopted by farmers (uses of compound, slow/controlled release, and organic and microbial fertilizers) and develop integrated agronomic and genetic strategies for Zn biofortification. A highly systematic and mechanistic model includes (a) migration paths of Zn (particularly from leaves to different grain parts) using isotopic labeling methods, (b) cross-talks between Zn and carbon, N, P, K, or other divalent cations, (c) inherent physiological and biochemical processes of enzymes and signaling phytohormones, and (d) complex genetic systems governing Zn homeostasis and their relationships with other nutrients, signaling molecules, and increase or dilution/penalty of yield under different environmental conditions (soil, water, and future climatic changes) and managements (breeding and

This is an open access article under the terms of the Creative Commons Attribution License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

© 2020 The Authors. *Food and Energy Security* published by John Wiley & Sons Ltd on behalf of Association of Applied Biologists

fertilization). These aspects require further elucidation to fully unravel the “black box” of Zn flow from source to sink.

KEYWORDS

biofortification, foliar sprays, isotopes, micronutrient, phytohormones, remobilization

1 | INTRODUCTION

Zinc (Zn) is an essential micronutrient for the health of plants, animals, and humans (Krężel & Maret, 2016; Marschner, 2012). It has been reported that more than 30% of the world's 6 billion population show Zn deficiency symptoms including retarded growth, anorexia, hypogeusia, diarrhea, pneumonia, pregnancy problems, and several other chronic diseases, especially in developing countries and rural areas (Brown, Peerson, Rivera, & Allen, 2002; Cakmak, 2008a; Das & Green, 2013; Ma et al., 2005; Ota et al., 2015; Prasad, 2013; United Nations International Children's Emergency Fund, 2012; White & Broadley, 2009). Together, Zn deficiency causes a 1.9% of the total global burden of disease (Rodgers et al., 2004). Wheat (*Triticum aestivum* L.) is one of the world's paramount cereal crops, providing up to 30% of human daily calorie intake (Cakmak, 2008a), and bread wheat alone is the staple food for 35% of the world's population (Poursarebani et al., 2014). However, wheat grains contain inherently low Zn, too low to meet daily human nutritional requirements, particularly when grown on Zn-deficient soils (Cakmak & Kutman, 2018). Furthermore, phytic acid (PA) and phenolic compounds act as antinutritional compounds in wheat grains and flour, which reduces the bioavailability of Zn in the human digestive tract (Gibson, Bailey, Gibbs, & Ferguson, 2010; Xia, Xue, Kong, et al., 2018; Xia, Xue, Liu, et al., 2018; Xue, Eagling, et al., 2014; Xue, Xia, McGrath, Shewry, & Zhao, 2015; Xue, Zhang, Liu, Xia, & Zou, 2016). The milling of wheat grains into white flours usually removes the Zn-rich parts (mainly the embryo and aleurone layer) retaining only the Zn-poor endosperm, which contains about 5–10 mg/kg Zn depending on the extraction rate, and cannot meet dietary Zn requirement (15–30 mg/kg in flour) (Brown, Hambridge, & Ranum, 2010; Cakmak, Pfeiffer, & McClafferty, 2010). It is therefore of great interest to biofortify wheat grain and flour with Zn to increase both concentration and bioavailability for human health benefits.

The concept of source and sink of biomass in crops was first proposed by Mason and Maskell (1928). It could also be applied to the study of Zn. The flow of not only photoassimilates, but also Zn, into developing grains (the sink) is determined by the source–sink relationship during the grain-filling period, and Zn may interact with photoassimilates (Liu, Zhang, Liu, Chen, & Zou, 2020). The Zn source is provided by both postanthesis Zn uptake and the remobilization of existing Zn reserves in vegetative organs (Rose &

Bowden, 2013). Sink capacity/strength is determined by the number and size of wheat grains and their ability to acquire Zn from source organs. The concentration of Zn in grain sink currently ranges between 20 and 35 mg/kg, at the global scale, with an average below or around 28–30 mg/kg. This concentration is well below the biofortification target range for human health, set by the HarvestPlus program (<http://www.harvestplus.org>) and FAO, which is usually 40–50 mg/kg (Allen, de Benoist, Dary, & Hurrell, 2006; Cakmak, Kalayci, et al., 2010; Chen, Zhang, et al., 2017; Graham et al., 2007; Hao, Zhang, & He, 2015; Ortiz-Monasterio et al., 2007; Pfeiffer & McClafferty, 2007; Tang et al., 2008; Wang, Mao, Zhao, Huang, & Wang, 2012). Conventional and molecular plant breeding, genetic engineering (transgenic technologies), and agronomic interventions including appropriate fertilizer applications are the major tools used to manipulate the source–sink relationship for the biofortification of wheat with Zn. It seems Zn biofortification of wheat can be achieved more rapidly by agronomic biofortification than by breeding or genetic modification (Cakmak & Kutman, 2018; Chen, Zhang, et al., 2017). Increasing source supply of Zn through soil and foliar fertilization can effectively improve grain Zn nutritional quality (concentration and bioavailability), when Zn partitioning and factors influencing sink growth and development remain unchanged. For example, in a field experiment involving 320 paired plots, an average increase in grain Zn of 10.5 mg/kg due to foliar Zn application was observed by Chen, Zhang, et al. (2017). Unfortunately, although much effort has been made, we are still far from fully understanding the source–sink interaction, and even further from rational manipulation. As a reflection of this uncertainty, breeders usually struggle with the dilution of Zn in wheat grain because of increases in yield. With the “Green Revolution” since the 1960s and improved soil and crop management strategies, the average yield of wheat has increased more than twofold (Curtis & Halford, 2014; Davis, 2009; Grassini, Eskridge, & Cassman, 2013; Tilman, Cassman, Matson, Naylor, & Polasky, 2002). However, these large increases in yield have commensurately caused considerable decreases in grain Zn concentrations, that is, the so-called dilution effect (Davis, 2009; Fan et al., 2008; Garvin, Welch, & Finley, 2006; Shewry, Pellny, & Lovegrove, 2016; Zhao et al., 2009). Furthermore, relationships between grain Zn concentrations and grain size or number of wheat grains are still unclear (Morgounov et al., 2007; Nowack, Schwyzer, & Schulin, 2008; Singh, Timsina, Lind, Cagno,

& Janssens, 2018; Velu, Ortiz-Monasterio, Cakmak, Hao, & Singh, 2014; Velu, Ortiz-Monasterio, Singh, & Payne, 2011; Velu et al., 2012; Xia, Xue, Liu, et al., 2018). Concurrently, biofortified genotypes newly developed by breeding or genetic modification are still unable to realize their full biological potential to acquire and accumulate the adequate target amount of Zn in their grains for a marked human health benefit, especially under “real-world” conditions without foliar or soil Zn fertilization, with low Zn availability in soils, and under various other environmental stresses such as drought and heat (Cakmak & Kutman, 2018). Additionally, the relative contributions of postanthesis Zn uptake by roots from soil and remobilization of Zn deposited in vegetative tissues (leaves and stems) to grain Zn accumulation differ between cultivars and even for the same cultivar under different conditions (Garnett & Graham, 2005; Kutman, Kutman, Ceylan, Ova, & Cakmak, 2012; Zhang et al., 2017). The source–sink relationship of these two sources may be affected by several plant and soil factors, including timing of senescence, length of the grain-filling period, the nutritional (e.g., nitrogen) status of the plant, and the Zn and water availability during grain filling.

The different source–sink relationships between different wheat cultivars/genotypes, or even within the same crop variety under different conditions, demand a mechanistic and systematic understanding of the source–sink interactions to achieve optimal Zn biofortification. Furthermore, the changing global climate (e.g., the elevated ambient air temperature and CO₂ concentration or the changed soil water availability due to altered precipitation) has exacerbated Zn deficiency in the soil–plant system in many countries and threatens human nutrition (Li, Ulfat, et al., 2019; Moreno-Jiménez et al., 2019; Myers et al., 2014). Understanding the source–sink relationship is therefore of great importance in facilitating wheat growth under stresses (Wang et al., 2019; Zhang et al., 2020), as demonstrated by Karim et al. (2012), who showed that foliar Zn spraying increased grain yield under drought, possibly by reducing the drought-induced oxidative cell damage due to improved antioxidative defense.

In this review, we apply the long-known concept of source and sink to wheat grain Zn biofortification for the first time. We summarize current observations and the effectiveness of manipulating the source–sink relationship for wheat grain Zn biofortification. We particularly focus on agronomic interventions and recapitulate what is known about the mechanisms involved in the translocation of Zn from soil to root, from root to shoot, and remobilization from vegetative tissues (especially leaves) to wheat grains. However, a systematic understanding of the source–sink interaction of Zn in wheat grains (especially in terms of molecular systems biology) and a well-coordinated source–sink relationship via a rational manipulation are required finally to realize the promised increase in grain Zn and crop yield on-farm. We also identify

unanswered questions and topics in need of further investigation to fully unravel the “black box” of Zn flow from source to sink.

2 | PHYSICAL MANIPULATION OF SOURCE/SINK BY DEFOLIATION, SPIKE SHADING, AND PARTIAL SPIKELET REMOVAL

Previous approaches for investigating the source–sink limitations of crop assimilates for grain growth and dry matter accumulation involve reducing the carbohydrate source from photosynthesis (e.g., through defoliation or spike shading) or reducing the grain sink size (e.g., through 50% spikelet removal) after anthesis (Austin & Edrich, 1975; Chang & Zhu, 2017). These approaches have also been applied to investigate the source–sink relationship of micronutrient accumulation in wheat grains (Xia, Xue, Kong, et al., 2018; Zhang, Zhang, et al., 2012; Zhang, Zhou, Zhang, & Wang, 2008). Zhang et al. (2008), Zhang, Zhang, et al. (2012) and Xia, Xue, Kong, et al. (2018) all reported experiments whereby defoliation, spike shading, and spikelet removal were employed to manipulate the source or sink of Zn and dry matter in wheat plants. Their results suggested that the accumulation of Zn and dry matter in grains is restricted by source supply and sink capacity, but the effects of reducing source supply or sink capacity on grain Zn concentrations are inconsistent: (a) Defoliation by removing all the leaf blades from tagged culms decreased the source-to-sink ratio, which led to decreases in grain Zn concentrations in the studies of Zhang et al. (2008) and Xia, Xue, Kong, et al. (2018), but increases in grain Zn concentrations in Zhang, Zhang, et al. (2012). (b) In experiments undertaken by Zhang et al. (2008), Zhang, Zhang, et al. (2012), spike shading and partial spikelet removal increased the source-to-sink ratio by reducing the grain number and size and correspondingly increased the grain Zn concentration, but Xia, Xue, Kong, et al. (2018) found that spike shading reduced grain Zn concentration.

Whether the Zn concentrations depend on the size of wheat grains and number of grains is also inconsistent in other studies (Morgounov et al., 2007; Nowack et al., 2008; Singh et al., 2018; Velu et al., 2011, 2012, 2014; Xia, Xue, Liu, et al., 2018). There seems to be no correlation between thousand grain weight and grain Zn in adapted wheat lines (Velu et al., 2012), indicating no concentration effect due to small grain size. However, there is a significant negative correlation in unadapted wheat (Morgounov et al., 2007), and a significant positive correlation found in our previous study (Xia, Xue, Liu, et al., 2018). Grain Zn concentrations were negatively correlated with grain numbers per m² (Morgounov et al., 2007), per pot (Singh et al., 2018), or spike numbers (Xia, Xue, Liu, et al., 2018), but no significant correlation

was observed between grain Zn concentrations and kernel numbers per spike (Xia, Xue, Liu, et al., 2018).

Such inconsistent and contradictory results are possibly due to different environmental conditions or the different genotypes of wheat varieties used. Zhang et al. (2008) tested nine different winter wheat cultivars, but Zhang, Zhang, et al. (2012) and Xia, Xue, Kong, et al. (2018), where exceptions to the trend were observed, used a single winter wheat cultivar “cv. Shijiazhuang 8” and “cv. Kenong 9204,” respectively. Therefore, the universality of the observations made by Zhang et al. (2008) needs to be verified collaboratively across multiple locations and multiple years and with more wheat varieties to identify the physiological and molecular regulatory mechanisms underpinning the above-mentioned impact of artificial source or sink manipulations on grain Zn accumulation and its interaction with the transport of photosynthates.

The flag leaf is the main photoassimilate source to grain filling, indicating the higher importance of the flag leaf than the other leaves to source–sink strength/relationship. Wheat flag leaf was found to contribute to more than 50% of grain filling, while its defoliation generated grain yield losses of 18%–30% (Wazzike, Yousfi, & Serghat, 2015). Other studies pointed out to the role of lower leaves that increases when the flag leaf area is affected by shading or defoliation (Wazzike et al., 2015). Similarly, the main source of micronutrients (including Zn) in the wheat grain is the flag leaf and, to a lesser extent, the lower leaves (Waters, Uauy, Dubcovsky, & Grusak, 2009). However, the effect of flag leaf defoliation on the source–sink relationship of grain Zn accumulation, the role of lower leaves affected by flag leaf defoliation, and the corresponding physiological and molecular mechanisms are unclear.

3 | INCREASING THE SOURCE OF EXOGENOUS ZN BY SOIL APPLICATION AND/OR FOLIAR SPRAYING

3.1 | Soil application

Application of Zn fertilizer to soil ensures sufficient uptake of Zn by wheat roots, prevents Zn deficiency symptoms, and improves grain yield and nutritional quality, especially Zn and protein concentration, particularly important for calcareous soils with low soil moisture and organic matter, and high pH (Cakmak, 2008a; Alloway, 2009; Cakmak & Kutman, 2018; Rengel, 2015). Field experiments have indicated that as the rate of Zn fertilizer applied to soil before sowing winter wheat increased gradually from 0 to 150 kg of $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ /ha, Zn concentrations in different milling fractions of wheat grain increased (Liu, Liu, Zhang, Chen,

& Zou, 2017). In addition, the total daily absorbed Zn (TAZ) by the human body (estimated Zn bioavailability using a trivariate model based on Zn homeostasis in the human intestine) and health impacts (as indicated by disability-adjusted life years [DALYs] saved) increased in whole flour, coarse flour, standard flour, and refined flour. Zn-biofortified standard and refined flour obtained with application of 50 kg of $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ /ha to the crop met the healthy diet requirement (3 mg bioavailable Zn obtained from 300 g wheat flour) and reduced the current health burden (DALYs) by >20% (Liu et al., 2017). Applying 300–1,500 kg of $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ /ha to soil at the wheat jointing stage significantly increased grain Zn concentrations from 31.9–46.0 to 58.0–71.6 mg/kg, while the molar ratios of phytic acid to Zn were significantly reduced from 26.2–30.7 to 12.9–17.3 (values below 15 indicating a good bioavailability) (Wang, Pan, et al., 2018). No additional benefits to grain Zn concentration and bioavailability were observed when the rates of Zn fertilizer ≥ 600 kg/ha were applied. 300 kg/ha had the maximum economic benefit due to lower costs associated with a lower Zn input (Wang, Pan, et al., 2018).

To correct or prevent Zn deficiency and thus to improve the yield in wheat plants (traditionally the main aim of Zn fertilization), rates of Zn applied to deficient soils usually range from 20 to 150 kg of $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ /ha (Abid, Ahmed, Qayyum, Shaaban, & Rashid, 2013; Cakmak, 2008b; Liu et al., 2017; Zhao, Tian, Cao, Lu, & Liu, 2014). Much more attention has been paid recently to biofortify wheat with Zn in the edible grains to improve human health outcomes since the start of the International HarvestPlus program. However, from an economic and environmental perspective, the optimal rate of Zn fertilizer applied to soil is not “the more the better.” Overuse of Zn, and the presence of contaminants such as Cd in low-quality Zn fertilizers, poses a risk of soil contamination and eventual toxicity to wheat plants and other organisms (Cakmak & Kutman, 2018; Liu, Liu, et al., 2020). A typical threshold concentration of Zn found in plant leaves, above which symptoms of toxicity are observed, is 300 mg/kg (Marschner, 2012). The optimal rate of soil Zn application and the thresholds above which Zn toxicity in soils and plant tissues occur depend on wheat cultivar, soil properties, and the timing and method of fertilization (broadcasting or banding) (Alloway, 2008; Liu, Liu, Zhang, Chen, & Zou, 2019; Marschner, 2012), which need to be studied accordingly.

3.2 | Foliar spraying

Besides the soil, there is another major source of Zn in the wheat grain: Zn deposited or absorbed in vegetative tissues (leaves and stems) can be remobilized and translocated into grains during the reproductive stage due to the high mobility of Zn in the phloem (Erenoglu, Nikolic, Römhild, & Cakmak, 2002; Erenoglu,

Kutman, Ceylan, Yildiz, & Cakmak, 2011; Sperotto, 2013; Waters et al., 2009). Many studies have demonstrated a positive correlation between the concentration of Zn in wheat grains and in leaves (Cakmak, Kalayci, et al., 2010; Haslett, Reid, & Rengel, 2001; Kutman, Yildiz, Ozturk, & Cakmak, 2010; Zou et al., 2012). Kutman et al. (2012) observed that remobilization of Zn from vegetative organs is critical for wheat grain Zn accumulation when Zn availability in soil is restricted during grain-filling period. Soils often have low moisture and organic matter contents in wheat cultivation areas (Alloway, 2009). Such adverse physical and chemical conditions considerably reduce the soluble Zn in the rhizosphere environment (Cakmak, 2008a), even after Zn fertilization of the soil. Therefore, providing a sufficient pool of physiologically available Zn within vegetative tissues (mainly leaves) by foliar spraying of Zn fertilizer effectively contributes to greater Zn deposition in wheat grains after flowering (Xia, Xue, Liu, et al., 2018).

Zn concentrations in all the organs of wheat (stem, leaf, grain, glume, and rachis) are significantly increased by foliar Zn spraying and increased with greater spraying doses (Zhang, 2017). However, the relationship between Zn fertilizer doses and increases in Zn concentrations in the wheat organs is not linear and foliar applications to wheat 10 days after flowering result in higher Zn tissue concentrations than applications made at flowering stage (Zhang, 2017). Therefore, the effectiveness of foliar Zn applications is related to the spraying location, timing, frequency, and rate (Cakmak, Kalayci, et al., 2010; Zhang, Sun, et al., 2012). Appropriate concentrations of foliar Zn spraying are generally in the range 0.1%–0.5% $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ (w/v) in water, but excessive concentrations could cause severe foliar damage and reduce yield (Cakmak, Kalayci, et al., 2010; Zhang, Sun, et al., 2012; Zhang et al., 2017). Cakmak, Kalayci, et al. (2010) found that the highest Zn concentrations in wheat grain are achieved by applying 0.5% $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ at each growth stage of stem + booting + milk + dough (four times in total), and a clear trend for later spraying (booting + milk) leading to higher grain Zn concentrations than earlier spraying. Zhang, Sun, et al. (2012) indicated that the spraying of 0.4% $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ (compared with 0.2% and 0.5%) at both the booting stage and 7 days after anthesis had the most significant effect. In general, differences observed may also relate to differences in soil type, the extent of soil Zn deficiency, the wheat variety used, and the climatic and environmental conditions at different experimental sites (Abdoli, Esfandiari, Mousavi, & Sadeghzadeh, 2014; Boonchuay, Cakmak, Rerkasem, & Prom-U-Thai, 2013; Welch, Graham, & Cakmak, 2013; Zhang et al., 2017).

3.3 | Foliar spraying versus soil application

Compared with soil Zn application, foliar Zn application is much more effective at increasing wheat grain and flour Zn concentration and bioavailability, leading to greater Zn

fertilizer use efficiency (Cakmak, Kalayci, et al., 2010; Cakmak, Pfeiffer, et al., 2010; Ozturk et al., 2006; Wang et al., 2012; Zhang, Shi, Rezaul, Zhang, & Zou, 2010; Zhang, Sun, et al., 2012). This may be due to the different routes of Zn movement to the developing seeds of wheat. The foliar-applied Zn can be directly transported from leaves to seeds via phloem loading and unloading, while the xylem discontinuity avoids direct Zn transport from roots to seeds and results in soil Zn first transferring from root to the shoot via the long-distance transport pathway in the xylem followed by the phloem before unloading into the grain (Erenoglu et al., 2002, 2011; Gupta, Ram, & Kumar, 2016; Haslett et al., 2001; Palmer et al., 2014). Moreover, mobility of Zn is higher in phloem than xylem due to increased concentration of chelating solutes (peptides, organic acids, etc.) in phloem sap (Gupta et al., 2016). A long-term soil fertilization trial whereby 15 kg/ha $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ was applied for 18 consecutive years on the Loess Plateau of China increased the grain yield of wheat from 2,364 to 2,464 kg/ha (an increase of only 4.2%) and increased the grain Zn concentration from 18.8 to 22.4 mg/kg (an increase of 19.2%) (Hao, Wei, & Dang, 2003). The grain Zn recovery of foliar spraying exceeded 26.4‰ and that of soil application was no more than 1.7‰ (Wang et al., 2012). In the HarvestZinc project, an international study covering seven countries (China, Kazakhstan, Pakistan, India, Turkey, Zambia, and Mexico), 23 experimental sites, 3 years, and 10 different wheat cultivars, an average 83.5% increase in grain Zn concentration was achieved by foliar Zn spraying, while soil Zn application at the time of sowing was less effective with an average increase of only 12% (Cakmak & Kutman, 2018; Zou et al., 2012).

In addition to its reliable adaptability and greater effectiveness in terms of biofortification of wheat grain with Zn without yield penalty, foliar Zn spraying is more economically efficient and has a lower environmental impact than soil Zn fertilization. A typical rate of Zn fertilizer applied as a foliar spray is usually only about 1 kg/ha or less (2–5 g/L $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$, ~23% of 500–1,000 L/ha), which is at least five times less than the lowest rate (5 kg Zn/ha) that is typically applied to soil (Boonchuay et al., 2013; Cakmak, Kalayci, et al., 2010; Ram et al., 2016). Applying foliar Zn fertilizer together with pesticides could reduce the cost of applying Zn, and there are no apparent compatibility issues (Ortiz-Monasterio, Cardenas, & Cakmak, 2015; Ram et al., 2016; Wang et al., 2015, 2016). Compared with separate foliar applications of Zn and insecticide, applications of insecticide/Zn mixtures could reduce the cost of application by 120 USD/ha and increase the net income of farmers by 6.3% (Wang et al., 2015).

There is uncertainty regarding whether wheat productivity is limited by the source or sink of photosynthates, or if it is limited by both factors (Chang & Zhu, 2017; Foulkes et al., 2011; Lv, Zhang, Zhang, Fan, & Kong, 2019). Similarly, inconsistent observations have been observed in

grain Zn accumulation of wheat. Zn concentrations in wheat grains are positively and linearly correlated with the rate of foliar Zn applied (Zhang, Sun, et al., 2012), indicating that the translocation of Zn from vegetative organs to grains is not a limiting factor and that grain Zn accumulation is most probably source limited. However, increasing the concentration of Zn in a culture solution from 10 to 100 $\mu\text{mol/L}$ failed to result in a corresponding 10-fold increase in the grain Zn concentration of wheat, perhaps as a result of saturating the membrane transporters during phloem loading (Pearson, Rengel, Jenner, & Graham, 1996). Stable isotope labeling in the ear culture system reveals two barriers for Zn transport into wheat grains: between the stem tissue rachis and the grain, and between the maternal and the filial tissue in the grain (Wang, Specht, & Horst, 2011). Therefore, the circumstances under which wheat grain Zn accumulation is limited by the source or the sink, and the detailed regulatory mechanisms responsible, require further study.

4 | SUPPLY OF EXOGENOUS CARBOHYDRATE, NITROGEN, PHOSPHORUS, AND OTHER NUTRIENTS, AND THEIR STATUS WITHIN THE PLANT AFFECTS GRAIN ZN ACCUMULATION OF WHEAT

4.1 | Carbohydrate

Exogenous carbohydrate supply or its status within the crop can influence the transport of Zn into the developing grains, and vice versa (Liu, Zhang, et al., 2020). The loading of Zn in phloem depends not only on the degree to which the membrane transporters are saturated, but also on the loading ability of sugars, amino acids, and macroelements (Grusak, Pearson, & Marentes, 1999). Pearson, Rengel, et al. (1996) observed that depletion of carbohydrate reserves in cultured wheat ears after maintaining them in darkness (prior to labeling) decreased the transport of radioisotope ^{65}Zn into grains, perhaps resulting from a decreased mass flow of carbohydrate in the phloem. Due to the limited grain sink capacity, sucrose, supplied at high amounts, may excessively accumulate in the peduncle and chaff, resulting in stomatal closure, the abatement of transportation via the xylem, and finally reduced Zn deposition in grains (Ma, MacKown, & Van Sanford, 1996). Several other studies observed that grain Zn concentration in cultured detached ears decreased with the increasing supply of sucrose, due to the increase in grain weight (i.e., a dilution effect) (Liu et al., 2014; Zhang, Zhang, et al., 2012). These observations may be due to the enhanced activities of enzymes involved in starch synthesis (Sasaki et al., 2005). Colabeling with ^{65}Zn and ^{14}C -sucrose revealed that Zn was

not transported within the grain in the same way as sucrose (Pearson, Jenner, Rengel, & Graham, 1996). ^{14}C -sucrose moves laterally out of the vascular system of the crease into the endosperm cavity and is subsequently taken up and stored in the endosperm. In contrast, ^{65}Zn appears to be retained within the vascular system of the crease and may be transported more slowly to grain parts such as the embryo and pericarp tissue. These above-mentioned observations were made on detached wheat ears. However, the effects of exogenous supply of sucrose (with or without Zn) on grain Zn accumulation of wheat grown in field conditions are unknown.

As early as in 1953, the addition of sucrose into urea sprays reduced the injury of maize leaves, perhaps by reducing the absorption rate and increasing the translocation rate of urea within the plant (Foy, Montenegro, & Barber, 1953). The impact of sucrose foliar sprays on grain Zn concentrations varies among different wheat cultivars and different environmental conditions (Xia, Xue, Kong, et al., 2018). Remarkably, a synergistic interaction was observed when spraying a combined foliar application of Zn and sucrose that resulted in improved wheat grain Zn concentration and bioavailability, compared to applications of Zn alone (Xia, Xue, Kong, et al., 2018; Xia, Xue, Kong, et al., 2018). Following Zhao et al. (2014), the synergistic impact of sucrose on Zn grain accumulation may be explained by the following three mechanisms: (a) a longer drying time of the mixed solution; (b) an improved penetration through the leaf cuticle; and (c) an enhanced rate of Zn translocation from the absorption site to the grain. As identified by Fernández et al. (2017), some additives to foliar sprays may lower the deliquescence relative humidity, thus promoting the rate of foliar absorption. Additionally, swelling of the cuticle due to absorption of substantial amounts of water may form “water-filled pores” (Schreiber, 2005), which benefit the penetration of hydrophilic solutes across the cuticle (Fernández et al., 2017). Therefore, sucrose may have played such roles, resulting in greater Zn absorption by the leaf. Further isotopic colabeling studies of $^{13/14}\text{C}$ -sucrose and $^{65/68}\text{Zn}$ - ZnSO_4 are required to ascertain whether the foliar-applied sucrose enters the leaf cells, accompanies, and facilitates the migration of exogenous and/or inherent Zn to grain, implicating cotransportation of carbohydrate and Zn through the plant.

4.2 | Nitrogen

In addition to the central role of nitrogen (N) fertilization in increasing wheat grain productivity, it also offers some benefits in terms of grain Zn biofortification. Interactions between N application and grain Zn nutrition have been investigated extensively on wheat. It has been found that zero or relatively low N supply results in low wheat grain Zn concentration, whereas optimal N supply achieves high grain Zn

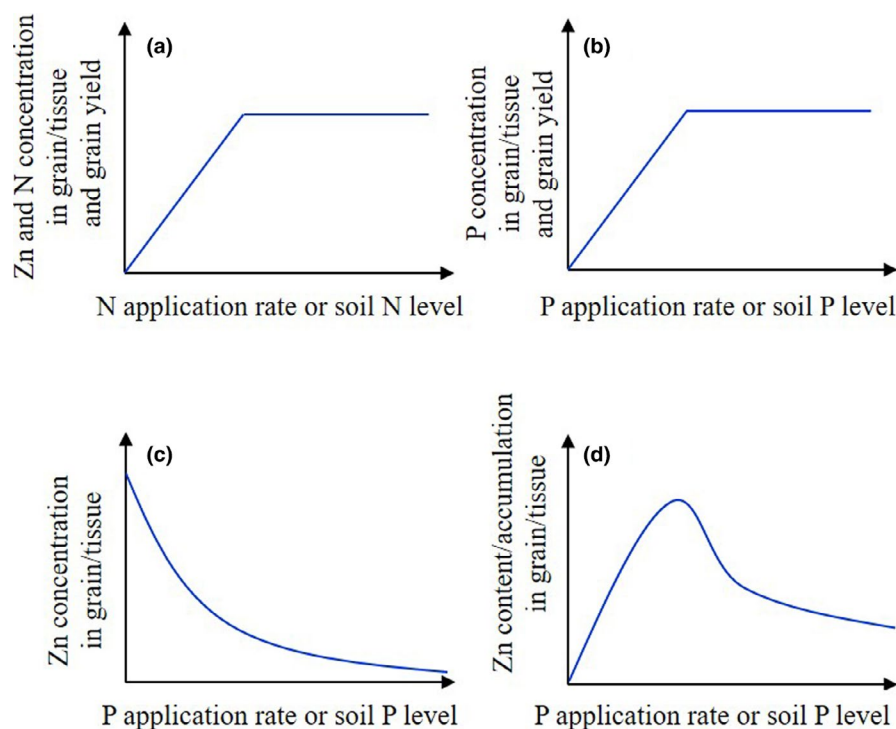
concentration, but excess N supply does not further increase grain Zn, grain N, or protein concentrations (Chen, Zhang, et al., 2017; Gooding, Fan, McGrath, Shewry, & Zhao, 2012; Kutman et al., 2010; Shi et al., 2010; Xia, Xue, Liu, et al., 2018; Xue et al., 2012; Xue, Zhang, et al., 2014; Figure 1). The use of radioisotope labeling (^{65}Zn) along with whole-plant partitioning revealed that optimal or adequate N supply not only improved Zn uptake by roots, but also the transport of Zn from roots to shoots and remobilization of Zn from leaves into wheat grains (Erenoglu et al., 2011; Kutman, Yildiz, & Cakmak, 2011; Xue, Zhang, et al., 2014). The reason is that appropriate N supply may promote the biosynthesis of nitrogenous Zn chelators or transporters, which play critical roles in Zn uptake, transport via xylem, and remobilization via phloem, as well as determining the wheat grain sink size for Zn (Chen, Zhang, et al., 2017; Kutman et al., 2010; Uauy, Distelfeld, Fahima, Blechl, & Dubcovsky, 2006). Analysis of the speciation and localization of Zn in wheat grains indicates that Zn interacts with proteins, and therefore, grain protein content (as a physiological sink for Zn) may determine the sink capacity (Cakmak, Pfeiffer, et al., 2010; Eagling et al., 2014; Ozturk et al., 2006; Persson et al., 2016; Xue, Eagling, et al., 2014; Xue, Zhang, et al., 2016). For example, protein and Zn staining showed colocalization of both within the grain of durum wheat, largely in the embryo and aleurone (Kutman et al., 2010). There was a significant positive correlation between concentrations of Zn and protein or N in wheat grain (Kutman et al., 2010; Xia, Xue, Liu, et al., 2018).

Clearly, optimized N management is required both from the perspective of yield improvement and environmental

protection (Cui et al., 2018). In practice, the “Action Plan for Zero Growth of Fertilizer Use by 2020” was formally issued by the Ministry of Agriculture of China in 2015 to reduce chemical fertilizer use nationally. Hence, China is undergoing a transformation from the overuse of N to a rational N input in crop production, and a 6-year field experiment with bread wheat (Chen, Zhang, et al., 2017) and our latest findings with four cultivars (Xia, Xue, Liu, et al., 2018) both highlight that such transformation will have no negative impacts on wheat grain yield or Zn concentration and bioavailability. However, N deficiency is still prevalent in parts of many developing countries (particularly those in sub-Saharan Africa), which may not only limit grain production, but also cause low grain nutritional quality (e.g., protein and Zn) (Abedi, Alemzadeh, & Kazemeini, 2011; Chen, Zhang, et al., 2017; Kutman et al., 2010; de Onis, Monteiro, Akre, & Clugston, 1993; Vitousek et al., 2009). Therefore, optimal N supply is necessary to sustain high grain yields to achieve food quantity security, and also to improve human dietary protein and Zn nutrition.

In addition to soil N application, foliar N spraying as urea has also been demonstrated to improve the Zn concentration of wheat grain (Kutman et al., 2010; Zhang, Sun, et al., 2012). Zn has a high mobility and can be easily translocated in the phloem tissue, and the remobilization of Zn from vegetative organs into seeds is an important mechanism for Zn deposition in the grain (Erenoglu et al., 2002; Haslett et al., 2001; Kutman et al., 2010). It seems that the exogenous foliar-applied N accelerates the route of inherent Zn from leaves to the grain; however, to the authors' knowledge, the specific migration footprint and underlying physiological, biochemical,

FIGURE 1 Conceptual curves illustrating the response of zinc (Zn) and nitrogen (N) concentration in grain/tissue and grain yield of wheat to N application rate or soil N level (a), the response of wheat grain yield and phosphorus (P) concentration to P application rate or soil P level (b), the response of Zn grain/tissue concentration to P application rate or soil P level (c), and the response of Zn content/accumulation in grain/tissue of wheat to P application rate or soil P level (d). The N or P application rate and their levels in the soil should be below the threshold harmful to wheat



and molecular mechanisms have never been investigated. Most previous studies focus on quantity effects of N, Zn, and their interactions on wheat grain yields and Zn nutrition (Liu et al., 2017; Xue, Eagling, et al., 2014; Xue, Eagling, et al., 2014; Xue, Xia, et al., 2016). However, very little attention has been paid to the influence of different chemical forms of N in fertilizers (e.g., urea-N, nitrate-N, ammonium-N), and the impacts of co-amendments applied to enhance N use efficiency (e.g., nitrification inhibitors, urease inhibitors, double inhibitors, and polymer-coated controlled release fertilizer) on grain Zn accumulation (Li et al., 2018; Zhang et al., 2013). This is a key area for future work.

4.3 | Phosphorus

Phosphorus (P) applied in excess of wheat requirements elevates plant-available P concentration in the soil, particularly after continuous P input for several crop seasons, and results in greater grain P, but a dramatic reduction in grain Zn concentration in wheat (Chen, Zhang, et al., 2017; Zhang et al., 2015, 2016; Zhang, Liu, Liu, Chen, & Zou, 2017; Zhang, Deng, et al., 2012). During year 2009–2012, a field experiment growing wheat was treated with various P rates (0, 25, 50, 100, 200, and 400 kg/ha calcium superphosphate) and demonstrated that, with an increase of soil Olsen-P from 3.4 to 40 mg/kg, grain Zn concentration declined from >30 to <15 mg/kg (Chen, Zhang, et al., 2017). Such phosphorus-induced reductions in the grain Zn concentration of wheat (i.e., density effect) may be caused by several processes, including (a) reduced plant availability of Zn due to precipitation as zinc phosphates, (b) decreased root exudation of organic anions that mobilize Zn, (c) decreased colonization of roots by arbuscular mycorrhizae (AM), (d) less translocation and remobilization of Zn from root and vegetative tissues to grains, and (e) the dilution effect induced by a yield increase (Hoffland, Wei, & Wissuwa, 2006; Mandal & Mandal, 1990; Ova, Kutman, Ozturk, & Cakmak, 2015; Teng et al., 2013; Yang, Tian, Lu, Cao, & Chen, 2011; Zhang et al., 2015, 2016; Zhang, Deng, et al., 2012). Furthermore, increasing soil P application rates also increases the phytate concentration in grain, coarse flour, standard flour, bread flour, and refined flour of wheat, thus decreasing Zn bioavailability (Zhang et al., 2017).

It appears the P-Zn antagonism mainly occurred between P and Zn concentrations within the wheat plant or between soil P application rates and wheat grain/shoot/root Zn concentrations. However, it does not always hold true for the relationship between soil P application rates and Zn contents/accumulation or remobilization in wheat (Zhang et al., 2015, 2016; Figure 1). With increasing P supply, Zn accumulation and Zn remobilization to grain, especially during

postanthesis, increased initially at low P application rates (25 and 50 kg P/ha), and then decreased gradually at higher rates (100–400 kg P/ha) (Zhang et al., 2015). Wheat grain yield and P concentration and accumulation increased with increasing P application rate, but then plateaued at higher rates (Chen et al., 2019; Zhang et al., 2015, 2016, 2017; Figure 1). Unlike the linear plateau model that applies to the relationship between P application rate and grain yield/shoot biomass/grain/shoot P accumulation of wheat, a single-peak curve may describe the tendency for root Zn uptake along with increasing P application rates (Figure 1). This curve also well describes the relationship between AM colonization and Zn accumulation in root and shoot of wheat at flowering stage, and in grain at maturity (Zhang et al., 2016). Therefore, the effect of P application rates on Zn concentration in wheat is different from that on Zn content/accumulation. P and Zn homeostasis within the wheat plant should both be further investigated to better understand such “concentration-content,” that is, “double C” relationships (Figure 1).

The P-Zn antagonism also occurs after foliar P spraying (Wang, Li, et al., 2018), which may on one hand result from reduced Zn penetration and absorption by leaves due to formation of phosphate precipitate in solution, on the surface of the leaf blade (Milani et al., 2012), or on the other hand result from a reduction in the physiological availability of Zn for translocation from leaves to grains via phloem (Cakmak & Marschner, 1987; Loneragan, Grove, Robson, & Snowball, 1979). Cakmak and Marschner (1987) indicated that sparingly soluble Zn-phosphate precipitates were the main reason why the physiological availability of Zn in cotton leaves decreased with increasing P supply.

During the last three decades, the available P concentrations in agricultural soils globally have dramatically increased, which may exacerbate Zn malnutrition (Alloway, 2008; Hotz & Brown, 2004; Li et al., 2011; MacDonald, Bennett, Potter, & Ramankutty, 2011). To achieve high wheat yields, the application of P is required. However, excessive P fertilization leads to surplus levels of P in croplands and eutrophication of water, and threatens human Zn nutrition (Chen, Zhang, et al., 2017; Cordell, Drangert, & White, 2009; Gemenet et al., 2015; MacDonald et al., 2011; Zhang et al., 2017). Therefore, optimal P management in intensive wheat production systems is needed to ensure high wheat yields, high levels of Zn in grain for human nutrition, and the protection of environmental quality. A systematic understanding of the physiological and molecular mechanisms responsible for plant uptake of P and Zn by roots from soil, their root-to-shoot translocation, and remobilization from vegetative tissues to grains under varying soil and environmental conditions is required for optimal P management, and improved genetic or agronomic Zn biofortification of wheat in the soil–crop–human continuum.

4.4 | Potassium

Combined foliar application of Zn and K has been shown to achieve greater uptake, concentration, and remobilization of Zn to wheat grain than spraying Zn alone (Wang, Li, et al., 2018). Moreover, a synergistic effect between K and Zn was observed by Srivastava, Ansari, Pachauri, and Tyagi (2016), in which soil applied K and soil/foliar-applied Zn increased the uptake of each other in grain, straw, and total uptake by wheat significantly, and thus enhanced the apparent utilization efficiency of each other. The facilitation effect of applied K on Zn absorption and translocation could be ascribed to its role in increasing (a) the leaf epidermis permeability and/or the stomatal activity for Zn penetration (Domnguez, Heredia-Guerrero, & Heredia, 2011; Roelfsema & Hedrich, 2005), (b) the nitrate reductase (NR) activity and the production of ATP leading to the efficient formation of N-containing molecules (e.g., NA, DMA, amino acids, and polypeptide) and transport of Zn through phloem to grain (Mohammad & Naseem, 2006), or (c) the sink size, that is, the synthesis of proteins in grain responsible for Zn storage (Mengel & Kirkby, 1987). Gao, Mohr, McLaren, and Grant (2011), however, found that potassium chloride (KCl) fertilizer increased wheat grain yield but concurrently decreased grain Zn concentration in 4 out of 6 site years, likely as a result of biological dilution. Therefore, there is no consensus on how Zn concentration and accumulation in wheat grains is affected by K application, which thus requires further investigation.

4.5 | Some other nutrients, including the Irving–Williams series metals

Irving–Williams divalent cations like Fe, Mn, Cu, Mg, Cd, Ni, and Co would compete with Zn for the same ligand-binding sites such as in phytosiderophores (PS), transporter proteins (like iron-regulated transporter 1), and nonselective cation channels (Gupta et al., 2016). Binding of Cd^{2+} , Pb^{2+} , Ni^{2+} , and Hg^{2+} ions to thiol groups of proteins causes protein inactivation and denaturation of various enzymes, and thus has detrimental effects on plant growth and also Zn accumulation, particularly in soils contaminated with such heavy metals (Gupta et al., 2016). Regarding Zn–Fe interactions, both metal ions have common transporter proteins required for their absorption by roots and transport to shoots (Gupta et al., 2016; Singh & Prasanna, 2020; Xue, Xia, et al., 2016). The concentrations of Zn and Fe in wheat grains were simultaneously increased by foliar Fe fertilization (Rakshit et al., 2016).

In terms of Se, Nawaz, Ahmad, Ashraf, Waraich, and Khan (2015) reported foliar Se application significantly increased shoot Se contents of wheat, whereas it reduced

Zn accumulation by 54% under drought stress. Xia, Yang, et al. (2019) reported the concentrations of Se and Zn in wheat grains were both elevated by foliar Se application. Interestingly, foliar application of Zn efficiently enhanced the wheat growth and Zn concentrations, and simultaneously alleviated wheat Cd toxicity and uptake from Cd-contaminated soil (Wu, Dun, Zhang, Li, & Wu, 2020). Simultaneous biofortification of wheat with Zn, I, Se, and Fe was obtained through foliar treatment of a micronutrient cocktail in six countries (Zou et al., 2019). Therefore, different elements influence the uptake and translocation each other, and result in antagonistic or synergistic effects. The impact of other nutrients on the source–sink relationships of Zn needs more extensive investigation and in-depth study.

5 | MOLECULAR MECHANISMS GOVERNING THE SOURCE–SINK FLOW OF ZN

5.1 | Uptake from soil and root-to-shoot translocation

Zn uptake and translocation are mainly influenced by the activities of transporter proteins located in the plasma membrane of root cells or the transporters participating in xylem loading and unloading (Erenoglu et al., 2011). At least six transporter families are involved in the uptake and/or transport of Zn, including Zn- and iron-regulated transporter-like proteins (ZIP), natural resistance-associated macrophage proteins (NRAMP), cation diffusion facilitator proteins (CDF), heavy metal ATPases (HMA), and yellow stripe-like (YSL) and ATP-binding cassette transporters. Among these, ZIP perform the function of Zn influx into the cytosol, are reported to facilitate Zn uptake by roots, and could also transport other metal ions (such as Fe^{2+} , Cu^{2+} , and Cd^{2+}) to the shoot (Guerinot, 2000; Ishimaru et al., 2005; Palmer & Guerinot, 2009). NRAMP also contribute to the transport of various divalent metal cations including Zn^{2+} , Fe^{2+} , Mn^{2+} , and Cu^{2+} (Nevo & Nelson, 2006). CDF are involved mainly in the sequestration of Zn into organelles (such as vacuoles, endoplasmic reticulum) or transfer the excess intracellular Zn^{2+} out of the cell when present at high Zn concentrations (Ricachenevsky, Menguer, Sperotto, Williams, & Fett, 2013). HMA participate in efflux of Zn to the apoplast and with YSL are responsible for xylem loading and unloading of Zn and other heavy metals (Curie et al., 2009; Hussain et al., 2004).

Multiple ZIP members have been identified in Arabidopsis, alfalfa, barley, grape, rice, soybean, and other plants (Nie et al., 2019). The expressions of ZIP genes in roots of several plant species are important in adaptation or response to Zn deficiency or fluctuations in soil

(Chen, Feng, & Chao, 2008; Grotz et al., 1998; Ishimaru et al., 2005; Li et al., 2013). Some of these genes are expressed in both epidermal/cortical and stellar cells in root, indicating potential roles in both Zn uptake and root-to-shoot translocation (Ishimaru et al., 2005; Milner, Seamon, Craft, & Kochian, 2013; Tiong et al., 2014, 2015). In wheat, little is known about this ZIP family with an exception of the higher expression of a Zn transporter *TdZIP1* from wild emmer wheat under Zn deficiency (Durmaz et al., 2011). A recent investigation using hydroponic and pot trials demonstrated that N supply enhanced Zn uptake and root-to-shoot translocation via up-regulating the expression of *TaZIP3* and *TaZIP7* in roots of winter wheat (*Triticum aestivum*) (Nie et al., 2019). The findings of Evens, Buchner, and Williams (2017) further demonstrated that the hexaploid wheat (*Triticum aestivum*) contains an expanded number of group F bZIP transcription factors that alter the expression of ZIPs by binding to Zn-deficiency-response elements in their promoters. The variation in particular conservation of cysteine-histidine-rich motifs throughout the wheat group F bZIPs and the corresponding spatial and temporal expression patterns of these genes may refine our understanding of the complex homeostatic network that serves in regulating Zn uptake and distribution during adaptation to low or changing Zn availability.

As strategy-II plant, roots of wheat can secrete mugineic acids (MA) or PS to chelate Zn^{2+} , which are subsequently taken up by roots through transporters (Gupta et al., 2020). Recently, twelve genes including nicotianamine synthase (*NAS1A*), *YSL6*, nicotianamine aminotransferase (*NAAT2D*), deoxymugineic acid synthase (*DMAS-1B*), 2'-deoxymugineic acid 2'-dioxxygenase, *NRAMP2*, zinc transporter 9-like, ferritin 1, mitoferrin-like 1, *ZIP5*, Fe^{2+} transporter protein 1-like (*IRT1*), and zinc induced facilitator-like 1 (*ZIF1*), involved in MA/PS biosynthesis and Fe/Zn uptake and transport, were identified in seedling and flag leaf, and showed higher expression in efficient wheat genotypes compared to inefficient genotypes for Fe and Zn accumulation (Gupta et al., 2020). miRNAs targeting the genes of Fe/Zn transportation and MA biosynthesis were also identified in this investigation (Gupta et al., 2020). These results will provide valuable resources for a better understanding of Zn transport and accumulation in wheat at the molecular level.

5.2 | Leaf senescence and remobilization

Nutrients and proteins in wheat grains are largely derived from the remobilization of degraded macromolecules during monocarpic leaf senescence, which is a genetically programmed and developmentally controlled active process characterized by large-scale coordinated changes in gene expression, catabolic activities, active transport, and

functional conduction (Cantu et al., 2011; Lim, Kim, & Nam, 2007; Woo, Kim, Lim, & Nam, 2019). NAC (NAM, ATAF1,2, CUC2) genes play important roles in plant developmental processes, auxin signaling, defense and abiotic stress responses, and leaf senescence (Lim et al., 2007; Woo et al., 2019). Uauy et al. (2006) reported the positional cloning of *Gpc-B1*, a wheat quantitative trait locus associated with increased protein, Zn, and Fe contents in grains. The ancestral wild emmer wheat allele encodes a NAC transcription factor (*NAM-B1*) that accelerates senescence and increases the remobilization of N, Zn, and Fe from leaves to developing grains. Five genes homologous to *TtNAM-B1* were isolated in common wheat. Depending on the homologous groups of chromosomes in which these genes reside, they are classified as *GPC-1* (*TaNAM-A1*, *TaNAM-B1*, and *TaNAM-D1*) and *GPC-2* (*TaNAM-B2* and *TaNAM-D2*) (Cantu et al., 2011; Wu, Dong, Yao, Zhao, & Gao, 2015). Most modern cultivated wheat varieties (i.e., 57 cultivated durum lines and a collection of 34 varieties of hexaploid wheat including the “Chinese Spring” and “Bobwhite”) lack the functional allele of *NAM-B1* or carry a nonfunctional *NAM-B1* allele due to a 1-bp frameshift insertion, only a few wheat varieties retain the wild-type *NAM-B1* allele (Uauy et al., 2006).

Uauy et al. (2006) further reduced the transcript/RNA levels of all the multiple *NAM* homologs using RNA interference (RNAi) to determine their effects on wheat development and nutrient remobilization. An RNAi construct was transformed into the hexaploid wheat variety “Bobwhite,” selected for its higher transformation efficiency relative to tetraploid wheat. A reduction in RNA levels of the *TaNAM* genes (*TaNAM-A1*, *TaNAM-B2*, *TaNAM-D1*, and *TaNAM-D2*) was associated with more than 3 weeks delay in whole-plant senescence, more than 30% decrease in grain protein, Zn, and Fe concentrations, and an increase in residual N, Zn, and Fe in the flag leaf, indicating the functional redundancy of these four genes and *TtNAM-B1*. To determine whether decreased remobilization, lower plant uptake, or decreased partitioning to grain are responsible for lower grain protein, Zn and Fe concentrations in the *NAM* knock-down line of “Bobwhite,” control and the RNAi wheat line with reduced expression of *NAM* genes were grown in potting mix and hydroponics when Fe or Zn was either withheld or provided in ample supply postanthesis. The results of Waters et al. (2009) suggest that a major effect of the *NAM* genes is an enhanced remobilization or efflux of nutrients from the vegetative tissues and a higher partitioning into grain, but not greater overall plant uptake. These above-mentioned researches indicate a central role for the *NAM* genes as transcriptional regulators of a direct link between the leaf senescence and nutrient remobilization to the developing wheat grain. Deciphering specific genes operating downstream of the *NAM* genes could provide new targets or a valuable entry

point to engineer more efficient efflux or remobilization of Fe, Zn, and N from source tissues to grain sink at the appropriate developmental stages (Waters et al., 2009).

A spatiotemporal expression pattern analysis of *NAM* transcription factors *Gpc-1* and *Gpc-2* in bread wheat cultivar “Chinese Spring” during grain filling showed, contrary to the observations of Uauy et al. (2006), that the functional *TaNAM-B1* rather than its dysfunctional paralog was found in “Chinese Spring,” and its nucleotide sequence was identical to the wild-type *TtNAM-B1* in *T. turgidum* var. *dicoccoides* (a wild emmer wheat) (Wu et al., 2015). All the results demonstrated that the no apical meristem (*NAM*) transcription factors *Gpc-1* (*TaNAM-A1*, *TaNAM-B1*, *TaNAM-D1*) and *Gpc-2* (*TaNAM-B2*, *TaNAM-D2*) were all widely expressed in studied tissues (penultimate leaf, flag leaf, peduncle, glume, rachis, and the kernel), with the exception in root where only the transcript of *Gpc-1* was observed. Wu et al. (2015) concluded that *Gpc-1* and *Gpc-2* were closely associated with mineral translocation in the grain. However, no obvious relationships between five *NAM* genes and the programmed cell death in wheat grains were found, and the discrepant temporal expression dynamics suggested that their functions were not identical. In addition, the potential effect of these genes on senescence in vegetative tissues was still elusive. Combined with previous findings, it is proposed that *Gpc-1* and *Gpc-2* may directly regulate nutrient remobilization alone or in parallel with senescence during grain filling (Wu et al., 2015).

The study of the transport mechanisms activated in the flag leaf of tetraploid wheat during monocarpic senescence confirms that *GPC1* is a key regulator of Zn remobilization which acts predominantly during the early stages of senescence (Pearce et al., 2014). *GPC1* has a clear role in the up-regulation of transmembrane transporter genes responsible for loading Zn into the phloem (*ZIP* and *YSL*) and in the up-regulation of genes involved in the biosynthesis of chelators (e.g., PS) that facilitate the phloem-based transport of Zn to the grains (*NAS*, *NAAT*) (Pearce et al., 2014).

Therefore, Zn remobilization and leaf senescence are intrinsically interconnected processes in most situations. Unfortunately, the gene regulatory networks controlling neither processes have been investigated under source–sink manipulations (e.g., foliar Zn spraying) nor elucidated to great depth. As Cantu et al. (2011) highlights, dedicated senescence libraries are absent in currently available wheat expressed sequence tag (EST) resources in the National Center for Biotechnology Information (NCBI). A better understanding of the gene regulatory networks is urgently required for further improvements in grain Zn nutritional value. The molecular mechanisms concerning the movement of Zn in different grain tissues such as embryo, scutellum, and aleurone layer need a further investigation.

6 | SIGNALING MOLECULES (PHYTOHORMONES) IN SOURCE–SINK INTERACTIONS

Phytohormones play important signaling roles in source–sink interaction. Various hormones coordinate responses to stress and the initiation and progression of monocarpic senescence in plants (Woo et al., 2019), with abscisic acid (ABA)-based chemical signaling playing an important role in promoting leaf senescence (Lim et al., 2007; Wilkinson & Davies, 2002; Woo et al., 2019). Exogenous ABA application induces senescence-associated mRNAs and accelerates leaf senescence (Lee et al., 2011). ABA levels appear to be elevated during senescence induced by a variety of biotic and abiotic stresses (e.g., drought or heat) (Sah, Reddy, & Li, 2016). Global gene expression analysis and characterization of genetic mutants have revealed that several other plant hormones potentially play roles in the promotion (e.g., jasmonic acid (JA), salicylic acid (SA), ethylene, and gibberellins (GAs, Chen, Xiang, Chen, Li, & Yu, 2017)) or suppression (e.g., cytokinins (Cortleven & Schmülling, 2015; Lim et al., 2007) and auxin (Cha et al., 2016; Feng, Xu, Wang, & Zhuoma, 2016; Kim et al., 2011; Lim et al., 2010)) of leaf senescence at all stages of leaf development (Woo et al., 2019). Melatonin improves the photosynthetic carbon assimilation and antioxidant capacity in wheat leaves exposed to Nano-ZnO stress (Zuo et al., 2017).

In addition to leaf development, ABA, ethylene, GAs, and cytokinins can greatly influence the wheat grain-filling process and Zn nutritional status. Generally, GA₃ and kinetin applications by seed soaking increased Zn contents in young barley plants; however, the highest Zn values in young wheat plants were obtained from the treatment that had no hormone (Akman, 2009). Yang, Zhang, Liu, Wang, and Liu (2006); Yang and Zhang (2018) found that elevated levels of endogenous ABA and higher ABA/ethylene and ABA/GAs ratios were required for the efficient filling of grain during the active wheat grain-filling period. Postanthesis moderate soil-drying or applying ABA at a low concentration elevated the ABA level, which can increase the activities of the key enzymes involved in carbohydrate metabolism in stem and grain, enhance assimilate loading and unloading capacity, and thereby accelerate transport and remobilization of assimilates to grains and promote starch synthesis in grain. Recent publications are showing clear impacts of manipulation of cytokinin dehydrogenase (CKX, the enzyme that inactivates cytokinin) in cereals on yield, root growth and orientation, and grain Zn nutrition (Chen, Zhao, Song, & Jameson, 2020). However, the direct relationship between CKX and Zn homeostasis genes/Zn-binding proteins in wheat plants has not been observed or established.

Since hormone signaling, root growth, leaf senescence, Zn remobilization, and kernel development are intrinsically linked together during the wheat grain-filling stage after anthesis (Cantu et al., 2011; Chen et al., 2020; Lim et al., 2007; Uauy et al., 2006; Woo et al., 2019; Wu et al., 2015; Yang & Zhang, 2018; Yang et al., 2006), the physiological and molecular mechanisms underlying their complex crosstalk need to be elucidated systematically to enable biofortification through efficient agronomic manipulation and agricultural improvement.

7 | GENETIC BIOFORTIFICATION OF WHEAT WITH ZN BY CONVENTIONAL AND MOLECULAR BREEDING AND GENETIC ENGINEERING

In addition to agronomic interventions, conventional and molecular breeding and genetic modification (transgenic technologies) are the other major tools available for improved biofortification of wheat with Zn (Cakmak & Kutman, 2018). Plant breeding represents the most economic and sustainable solution to malnutrition problems by exploiting natural genetic variation to produce new Zn-enriched wheat varieties (Sharma, Aggarwal, & Kaur, 2017; Stein, 2010). Substantial genetic variation in grain Zn densities among different wheat germplasms has been observed and exploited for breeding in recent years in the HarvestZinc project (Chen, Zhang, et al., 2017; www.harvestzinc.org). Published data from multiple field trials show that an increase of grain Zn by 6.5–10 mg/kg could be achieved by elite lines compared to local controls, indicating a marked biological effect in target populations and promising progress in breeding and bringing Zn-enriched wheat varieties to market (Bouis, Hotz, McClafferty, Meenakshi, & Pfeiffer, 2011; Chen, Zhang, et al., 2017; Velu et al., 2014). However, the development of new genotypes by breeding or genetic engineering also risks drawbacks, such as yield trade-off, the inability to realize their full potential and unstable traits of grain Zn under different soils and environmental conditions, particularly with inadequate soil Zn availability (Cakmak & Kutman, 2018; Stein, 2010).

Transgenic research is still in its infancy, and there are very few encouraging examples of transgenic wheat plants developed for improved root uptake, transport, and grain accretion capacity for Zn (Borrill et al., 2014). Genetically modified plants are often tested in controlled environments, mostly with sufficient Zn availability, and lack performance verification under natural field conditions with limited Zn availability alongside various other stresses such as drought, heat, and disease. As Cakmak and Kutman (2018) state: “what purpose can an upgraded transport and storage system

serve if the amount of goods to be transported and stored is limited anyway?”

Looking to the future, a range of emerging proteomic, metabolomic, transcriptomic, and epigenomic studies and genome-wide association studies (GWAS) for investigating Zn uptake, Zn transport, and Zn grain deposition to identify the bottlenecks of Zn biofortification in wheat would greatly improve our current understanding on the Zn homeostasis network. These investigations would undoubtedly contribute to molecular design, molecular-assisted labeling, and transgenic breeding, which can increase the selection efficiency and breeding accuracy and shorten the breeding duration. Grain Zn accumulation involves the spatiotemporal regulation of complex networks, and there are rapidly expanding genetic resources and knowledge for manipulating Zn homeostasis in wheat. Therefore, a finer approach, such as the emerging hot CRISPR/Cas9 system for genome editing, should help accelerate advances in Zn biofortification.

8 | IMPACTS AND MITIGATION OF CLIMATIC CHANGE

Moreno-Jiménez et al. (2019) suggest that the increased aridity driven by climate change (rising temperatures, less rainfall, and lower water availability) will indirectly limit the availability of essential micronutrients for organisms, particularly Fe and Zn, by increasing soil pH and decreasing soil organic matter in drylands. These negative impacts may pose serious threats to key ecological processes and services, compromising global food security. However, zinc fertilizer is known to alleviate the negative effects of high-temperature stress on flag leaf nitrate reductase and glutamine synthetase activities, and on grain and protein yields of wheat (Tao et al., 2018). Furthermore, foliar spraying of Zn improved the grain yield of wheat under drought conditions, even in a soil with high DTPA-extractable Zn (Karim et al., 2012). This is possibly because the extra Zn supplemented the Zn requirement of wheat plants, enhances antioxidative defenses, and thus decreases the drought-induced oxidative cell damage (Cakmak, 2000).

Multigenerational exposure to elevated atmospheric CO₂ could enhance grain yields but reduce grain N, K, calcium (Ca), protein, glutenin macropolymer, and total amino acid concentrations in wheat (Li, Ulfat, et al., 2019). Myers et al. (2014) reported that C₃ (e.g., wheat, barley, rice) grains had lower Zn concentrations when grown under field conditions at elevated atmospheric CO₂ concentration, whereas C₄ crops (e.g., maize) seemed to be less affected, due to their different physiology. A relatively greater amount of CO₂ concentrating internally in C₄ crops, compared to C₃ crops, may lead to photosynthesis

being CO₂-saturated even under ambient CO₂ conditions, thus resulting in no stimulation of photosynthetic carbon assimilation at elevated CO₂ levels under mesic growing conditions (Leakey, 2009). Therefore, the greater effectiveness of spraying foliar “sucrose + Zn” rather than Zn only in improving Zn concentration and bioavailability in wheat (Xia, Xue, Kong, et al., 2018; Xia, Xue, Kong, et al., 2018), but not in maize grains (Xia, Kong, et al., 2019), may also be related to the different physiology of these two crops. As C₄ maize has a higher photosynthetic capacity to produce more carboxylates than C₃ wheat, its grain mineral accumulation is thus less dependent on an exogenous foliar sucrose supply. Differences between different cultivars of a single crop suggest that breeding for decreased sensitivity to increasing atmospheric CO₂ could partly address the emerging threat of climate change to human nutrition (Myers et al., 2014).

Unfortunately, the impacts of climatic change on the source–sink relationship of Zn in wheat plants and their adaption strategy have been much less investigated. These impacts must be considered when designing sustainable or climate-smart management actions (including breeding) to achieve the goal of biofortification, and to fully understand how forecasted scenarios of climatic change will affect the agricultural and nutritional potential of wheat worldwide. Drought and heat stresses, elevated atmospheric CO₂ concentration, and their interactive effects, influence wheat growth parameters (such as the stomatal conductance, plant water relation, ABA concentrations in leaf and spike, photosynthesis, leaf senescence, and grain yield components) (Li, Kristiansen, Rosenqvist, & Liu, 2019; Li, Li, et al., 2019; Li, Ulfat, et al., 2019). However, it is unclear how these parameters may affect the source–sink relationship of Zn, and what the underlying physiological, biochemical, and molecular mechanisms are.

9 | CONCLUSIONS AND FUTURE DIRECTIONS

9.1 | Integrated strategies for zinc biofortification

Different results have been obtained for Zn biofortification in wheat grains from different experiments. In some experiments, higher soil DTPA-extractable Zn concentrations did not lead to higher wheat grain Zn concentration (Cakmak, Kalayci, et al., 2010). Other experiments reveal a large genotypic variation in wheat grain Zn concentrations (Gomez-Coronado, Poblaciones, Almeida, & Cakmak, 2016). The increases in grain Zn concentration after foliar Zn spraying varied widely from 14.9% to 106.9% (Cakmak, Kalayci, et al., 2010; Wang et al., 2012; Zhang, Sun, et al., 2012;

Zou et al., 2012), with even greater increases (from 12–20 to 42–52 mg/kg) recorded by Gomez-Coronado et al., (2016). It appears that while foliar Zn spraying is more effective than soil N or Zn application to enrich wheat grains with Zn, the grain Zn bioavailability is influenced more by cultivar selection (Xia, Xue, Liu, et al., 2018). Trends in land management and environmental change may decrease future wheat grain Zn accumulation due to adverse soil and climatic factors including inadequate N use or overuse of P, depleting the already low soil available Zn and drought prevalence, and increasing atmospheric CO₂ concentrations (Chen, Zhang, et al., 2017).

In addition to the need to improve wheat grain Zn nutritional quality, there is also a need to produce grain with a similar yield and mineral concentration using less fertilizer (e.g., Zn, N, and P) and other exogenous inputs to obtain economic and environmental benefits. No single intervention or strategy may work alone to effectively achieve the biofortification target. Integrative strategies are particularly needed, in which all factors, including cultivars, soil type, environmental conditions, and agronomy (e.g., soil and foliar fertilization), are considered and managed holistically. Several case studies have shown a clear potential for an integrated strategy. Zhang, Deng, et al. (2012) indicated that the reduction in Zn concentration in wheat grain due to increased P fertilization could be mitigated by foliar Zn application. A combination of “Zn + N” or “Zn + K” spraying was more effective than Zn alone for enrichment of wheat grains with Zn (Wang, Li, et al., 2018). Xia, Xue, Liu, et al. (2018) also showed that optimal soil N and foliar Zn management together with suitable wheat cultivars maintained high grain yield with lower N input and simultaneously substantially increased the Zn concentration, bioavailability, and thus the nutritional quality of whole flours.

Here, we propose a conceptual framework for an integrated strategy to maximize grain Zn nutritional quality while ensuring high yields and protecting the environment. At least four factors should be managed in coordination in a sustainable and intensified wheat production system: (a) adoption of biofortified cultivars with low grain phytate and high grain Zn concentration, high yield, and resistance to biotic and abiotic stresses; (b) creation of an adequate available Zn pool in wheat shoots for Zn retranslocation to grain during post-anthesis via efficient foliar Zn spraying in combination with soil Zn application based on the initial soil DTPA-extractable Zn status prior to sowing; (c) optimization of N fertilizer application rates that ensure optimal grain yield, greater grain Zn concentrations, and lower N losses; and (d) avoiding the overuse of P fertilizer by maintaining soil available P at appropriate levels that meet crop requirements but prevent P-Zn antagonism and environmental pollution. Using our strategy, the target for biofortification will be rapidly achieved by combining agronomic and genetic strategies. The development of

a molecular systems model of source–sink interactions (linking the molecular mechanisms to physiological functions and the outcome of grain Zn status) is required to develop a strategy to rationally manipulate the source–sink relationship for Zn biofortification of wheat based on specific requirements (Chang & Zhu, 2017).

9.2 | Greater focus on farmers' fertilization practice

The development of basic concepts and theory of Zn nutrition in wheat plants has resulted in a separation between experimental work and the practices that farmers adopt when they apply fertilizer. In many studies, the frequency of foliar spraying is at least 2 or 3 times, which creates additional labor costs for farmers. Furthermore, farmer adoption of experimental treatments is hampered by the lack of newly developed high-efficient foliar Zn fertilizers that are unavailable to farmers. Much more attention has been paid to the

dose effects of a single element (Zn, N, or P), but not on their chemical forms or the interactions between these elements. There is a lack of research on compound, slow/controlled release, and organic and microbial fertilizers. Therefore, the development of new types of foliar Zn fertilizers and research on the efficacy of compound, slow/controlled release, and organic and microbial fertilizers on Zn nutrition in wheat plants should be strengthened in the very near future.

9.3 | Unraveling the uncertain fate of foliar-applied Zn

There is abundant evidence showing that foliar Zn spraying has great potential for improving grain Zn nutritional quality of wheat. However, our current understanding of the fate of foliar-applied Zn from wheat surface to metabolism and the factors that influence the ultimate efficacy of foliar Zn application is uncertain and incomplete (Fernández & Brown, 2013). In wheat, significant Zn mobility can occur

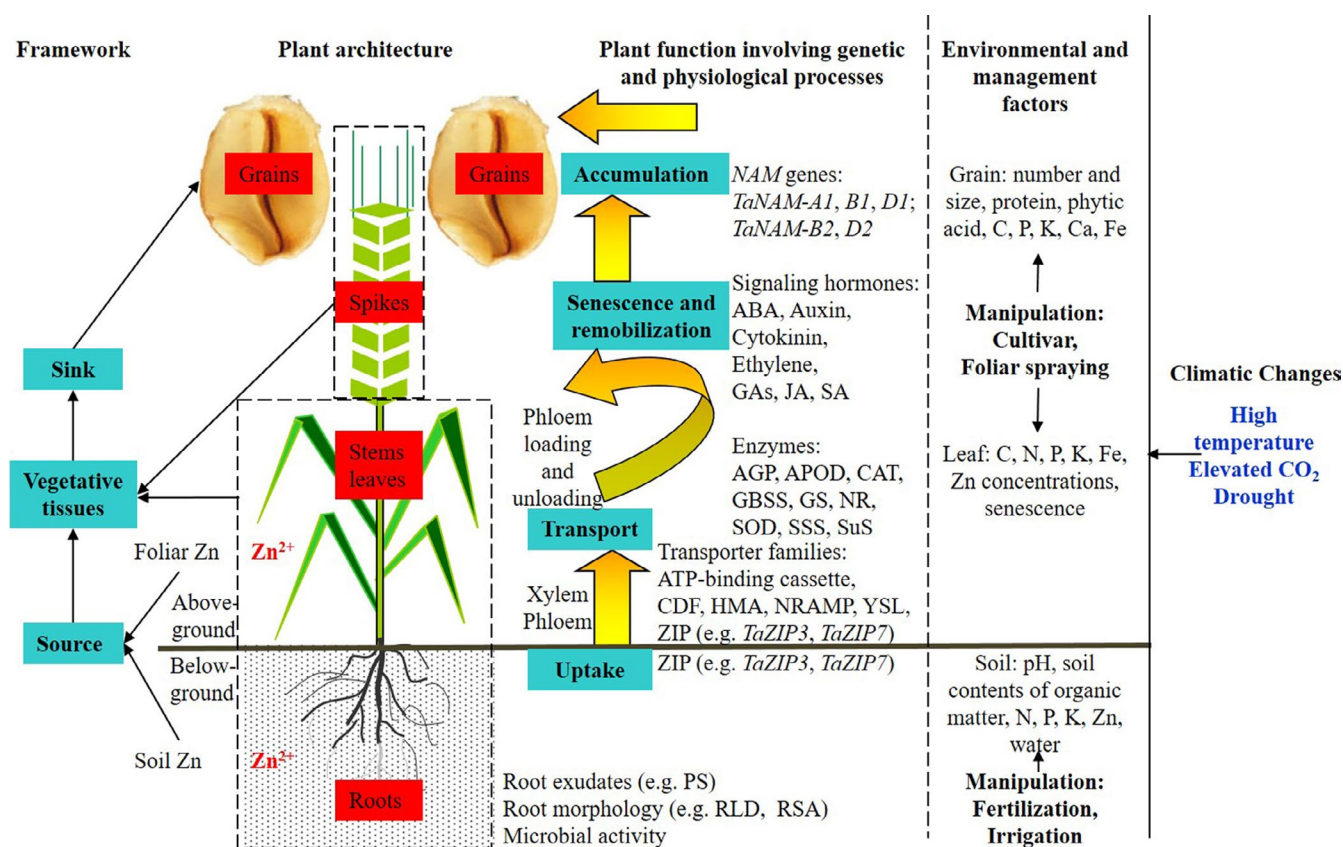


FIGURE 2 A schematic diagram of the proposed framework of a mechanistic model representing source–sink relationships of Zn in wheat grains, involving genetic and physiological processes and environmental and management factors. ABA, abscisic acid; AGP, adenosine diphosphate (ADP)-glucose pyrophosphorylase; APOD, ascorbate peroxidase; ATP, adenosine triphosphate; C, carbon; Ca, calcium; CAT, catalase; CDF, cation diffusion facilitator proteins; Fe, iron; GAs, gibberellins; GBSS, granule-bound starch synthase; GS, glutamine synthetase; HMA, heavy metal ATPases; JA, jasmonic acid; K, potassium; N, nitrogen; NAM, no apical meristem; NR, nitrate reductase; NRAMP, natural resistance-associated macrophage proteins; P, phosphorus; PS, phytosiderophores; RLD, root length density; RSD, root surface area; SA, salicylic acid; SOD, superoxide dismutase; SSS, soluble starch synthase; SuS, sucrose synthase; Ta, triticum aestivum; YSL, yellow stripe-like transporters; ZIP, zinc- and iron-regulated transporter-like proteins; Zn, zinc

but it is strongly dependent on factors such as plant nutritional status or plant phenological state determining the import or export of Zn (Erenoglu et al., 2011; Fernández & Brown, 2013; Kutman et al., 2012; Lim et al., 2007; Woo et al., 2019). Specifically, the pathway of the exogenous Zn penetrating the wheat epidermis (the cuticle, the epidermal cells including trichomes or stomata), the apoplast, the symplast, and finally transport from leaf to sink grain, and the underlying molecular, physiological, and biochemical mechanisms, need to be elucidated systematically. Use of radioisotopes (^{65}Zn) or stable isotopes (^{68}Zn) is a promising technology to track the movement of Zn within wheat plants (Erenoglu et al., 2011; Xue et al., 2015). Here, we suggest that colabeling of foliar-applied $^{65/68}\text{Zn}$, $^{13/14}\text{C}$, ^{15}N , and even ^{32}P in combination with hormone signaling and a series of omics analysis including proteome, metabolome, transcriptome, genome, and ionome is required to fully unravel the uncertain fate of foliar-applied Zn and influencing factors, especially during leaf senescence of wheat. A sound understanding of the complex mechanisms responsible for the ultimate delivery of foliar-applied Zn to sink grain involves anatomical, physicochemical, biological, and environmental principles that govern the absorption, translocation, and utilization of foliar-applied Zn by wheat, and this understanding is essential for improving the effectiveness and performance of foliar Zn fertilizers.

9.4 | Toward a mechanistic model of source–sink interactions of Zn in wheat

A mechanistic model of source–sink interactions of Zn in wheat could be developed to include modules for source (vegetative tissues), sink (grains), and the related transport processes (flow via xylem and phloem) (Figure 2). At a minimum, detailed Zn metabolic processes in different organs (roots, leaves, and grains), the transport processes in stem and between organs, and the architecture of wheat plants need to be considered in this model. Preexisting models could be adapted or used to define the framework of a new model of Zn source–sink interactions. Examples of such models include the C and N metabolism model in root, temporary storage pool, leaf, and seeds/fruits, the short-distance or long-distance transport model, and the 3D shoot or root architecture model (Chang & Zhu, 2017). The complex genetic control system regulating the source–sink flow of Zn (including uptake from soil, root-to-shoot translocation, and leaf senescence and remobilization) and its response and adaptation to various environmental conditions (e.g., soil type, water availability, climatic conditions) and management tools (e.g., breeding and fertilization) should be the core of this mechanistic model (Figure 2). From inside out, the model framework should establish the relationships

between inner genes (G) regulating Zn homeostasis in the wheat plant and outer influencing factors, such as the environment (E) and management (M). The model should also link molecular mechanisms to physiological functions and the outcome of grain Zn status. With a greater understanding of $G \times E \times M$ interactions, more available knowledge can be incorporated and the model updated to fully represent the “black box” of the source–sink relationships of Zn in wheat.

ACKNOWLEDGMENTS

This research was financially supported by the Shandong Provincial Key Research and Development Program of China (2018GNC111012), the State Key Laboratory of Crop Biology of China (2016KF05), the National Key Research and Development Program of China (2016YFD0300202, 2017YFD0301005, 2018YFD0200603), the National Natural Science Foundation of China (31702001), the Shandong Provincial Innovation Project for Agricultural Key Technology Application: Research and Demonstration of Annual Planting Patterns and Key Technologies for Green Improvement of Grain, Industrial and Forage Crops Production, and the Innovation Project of Shandong Academy of Agricultural Sciences of China (CXGC2016B04).

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

ORCID

Haiyong Xia  <https://orcid.org/0000-0002-6806-0507>

REFERENCES

- Abdoli, M., Esfandiari, E., Mousavi, S. B., & Sadeghzadeh, B. (2014). Effects of foliar application of zinc sulfate at different phenological stages on yield formation and grain zinc content of bread wheat (cv. Kohdasht). *Azarian Journal of Agriculture*, 1, 11–16.
- Abedi, T., Alemzadeh, A., & Kazemeini, S. A. (2011). Wheat yield and grain protein response to nitrogen amount and timing. *Australian Journal of Crop Science*, 5(3), 330–336. <https://doi.org/10.2134/agronj2010.0325>
- Abid, M., Ahmed, N., Qayyum, M. F., Shaaban, M., & Rashid, A. (2013). Residual and cumulative effect of fertilizer zinc applied in wheat-cotton production system in an irrigated arid soil. *Plant, Soil and Environment*, 69, 505–510. <https://doi.org/10.17221/313/2013-PSE>
- Akman, Z. (2009). Effect of plant growth regulators on nutrient content of young wheat and barley plants under saline conditions. *Journal of Animal and Veterinary Advances*, 8(10), 2018–2021.
- Allen, L., de Benoist, B., Dary, O., & Hurrell, R. (2006). *Guidelines on food fortification with micronutrients*. Geneva, Switzerland: WHO.
- Alloway, B. J. (2008). *Zinc in soils and crop nutrition* (2nd ed.). Brussels, Belgium: International Zinc Association and International Fertilizer Industry Association.
- Alloway, B. J. (2009). Soil factors associated with zinc deficiency in crops and humans. *Environmental Geochemistry and Health*, 31(5), 537–548. <https://doi.org/10.1007/s10653-009-9255-4>

- Austin, R. B., & Edrich, J. (1975). Effects of ear removal on photosynthesis, carbohydrate accumulation and on the distribution of assimilated ^{14}C in wheat. *Annals of Botany*, 39(2), 141–152. <https://doi.org/10.1093/oxfordjournals.aob.a084922>
- Boonchuay, P., Cakmak, I., Rerkasem, B., & Prom-U-Thai, C. (2013). Effect of different foliar zinc application at different growth stages on seed zinc concentration and its impact on seedling vigor in rice. *Soil Science and Plant Nutrition*, 59, 180–188. <https://doi.org/10.1080/00380768.2013.763382>
- Borrill, P., Connorton, J. M., Balk, J., Miller, A. J., Sanders, D., & Uauy, C. (2014). Biofortification of wheat grain with iron and zinc: Integrating novel genomic resources and knowledge from model crops. *Frontiers in Plant Science*, 5, 53. <https://doi.org/10.3389/fpls.2014.00053>
- Bouis, H. E., Hotz, C., McClafferty, B., Meenakshi, J. V., & Pfeiffer, W. H. (2011). Biofortification: A new tool to reduce micronutrient malnutrition. *Food and Nutrition Bulletin*, 32(1_suppl1), S31–S40. <https://doi.org/10.1177/15648265110321S105>
- Brown, K. H., Hambridge, K. M., & Ranum, P. (2010). Zinc fortification of cereal flours: Current recommendations and research needs. *Food and Nutrition Bulletin*, 31(1_suppl1), S62–S74. <https://doi.org/10.1177/15648265100311s106>
- Brown, K. H., Pearson, J. M., Rivera, J., & Allen, L. H. (2002). Effect of supplemental zinc on the growth and serum zinc concentrations of prepubertal children: A meta-analysis of randomized controlled trials. *American Journal of Clinical Nutrition*, 75(6), 1062–1071. <https://doi.org/10.1093/ajcn/75.6.1062>
- Cakmak, I. (2000). Possible roles of zinc in protecting plant cells from damage by reactive oxygen species. *New Phytologist*, 146(2), 185–205. <https://doi.org/10.1046/j.1469-8137.2000.00630.x>
- Cakmak, I. (2008a). Enrichment of cereal grains with zinc: Agronomic or genetic biofortification? *Plant and Soil*, 302(1–2), 1–17. <https://doi.org/10.1007/s11104-007-9466-3>
- Cakmak, I. (2008b). Zinc deficiency in wheat in Turkey. In B. J. Alloway (Ed.), *Micronutrient deficiencies in global crop production* (pp. 181–200). Dordrecht, The Netherlands: Springer Science + Business Media.
- Cakmak, I., Kalayci, M., Kaya, Y., Torun, A. A., Aydin, N., Wang, Y., ... Horst, W. J. (2010). Biofortification and localization of zinc in wheat grain. *Journal of Agricultural and Food Chemistry*, 58(16), 9092–9102. <https://doi.org/10.1021/jf101197h>
- Cakmak, I., & Kutman, U. B. (2018). Agronomic biofortification of cereals with zinc: A review. *European Journal of Soil Science*, 69(1), 172–180. <https://doi.org/10.1111/ejss.12437>
- Cakmak, I., & Marschner, H. (1987). Mechanism of phosphorus-induced zinc deficiency in cotton. III. Changes in physiological availability of zinc in plants. *Physiologia Plantarum*, 70(1), 13–20. <https://doi.org/10.1111/j.1399-3054.1987.tb08690.x>
- Cakmak, I., Pfeiffer, W. H., & McClafferty, B. (2010). Biofortification of durum wheat with zinc and iron. *Cereal Chemistry*, 87(1), 10–20. <https://doi.org/10.1094/CCHEM-87-1-0010>
- Cantu, D., Pearce, S. P., Distelfeld, A., Christiansen, M. W., Uauy, C., Akhunov, E., ... Dubcovsky, J. (2011). Effect of the down-regulation of the high Grain Protein Content (GPC) genes on the wheat transcriptome during monocarpic senescence. *BMC Genomics*, 12, 492. <https://doi.org/10.1186/1471-2164-12-492>
- Cha, J.-Y., Kim, M. R., Jung, I. J., Kang, S. B., Park, H. J., Kim, M. G., ... Kim, W.-Y. (2016). The thiol reductase activity of YUCCA6 mediates delayed leaf senescence by regulating genes involved in auxin redistribution. *Frontiers in Plant Science*, 7, 626. <https://doi.org/10.3389/fpls.2016.00626>
- Chang, T.-G., & Zhu, X.-G. (2017). Source-sink interaction: A century old concept under the light of modern molecular systems biology. *Journal of Experimental Botany*, 68(16), 4417–4431. <https://doi.org/10.1093/jxb/erx002>
- Chen, L., Xiang, S., Chen, Y., Li, D., & Yu, D. (2017). *Arabidopsis* WRKY45 interacts with the DELLA protein RGL1 to positively regulate age-triggered leaf senescence. *Molecular Plant*, 10(9), 1174–1189. <https://doi.org/10.1016/j.molp.2017.07.008>
- Chen, L., Zhao, J., Song, J., & Jameson, P. E. (2020). Cytokinin dehydrogenases: A genetic target for yield improvement in wheat. *Plant Biotechnology Journal*, 18(3), 614–630. <https://doi.org/10.1111/pbi.13305>
- Chen, W., Feng, Y., & Chao, Y. (2008). Genomic analysis and expression pattern of *OsZIP1*, *OsZIP3*, and *OsZIP4* in two rice (*Oryza sativa* L.) genotypes with different zinc efficiency. *Russian Journal of Plant Physiology*, 55(3), 400–409. <https://doi.org/10.1134/s1021443708030175>
- Chen, X.-X., Zhang, W., Liang, X.-Y., Liu, Y.-M., Xu, S.-J., Zhao, Q.-Y., ... Zou, C.-Q. (2019). Physiological and developmental traits associated with the grain yield of winter wheat as affected by phosphorus fertilizer management. *Scientific Reports*, 9, 16580. <https://doi.org/10.1038/s41598-019-53000-z>
- Chen, X.-P., Zhang, Y.-Q., Tong, Y.-P., Xue, Y.-F., Liu, D.-Y., Zhang, W., ... Zou, C.-Q. (2017). Harvesting more grain zinc of wheat for human health. *Scientific Reports*, 7, 7016. <https://doi.org/10.1038/s41598-017-07484-2>
- Cordell, D., Drangert, J.-O., & White, S. (2009). The story of phosphorus: Global food security and food for thought. *Global Environmental Change*, 19(2), 292–305. <https://doi.org/10.1016/j.gloenvcha.2008.10.009>
- Cortleven, A., & Schmölling, T. (2015). Regulation of chloroplast development and function by cytokinin. *Journal of Experimental Botany*, 66(16), 4999–5013. <https://doi.org/10.1093/jxb/erv132>
- Cui, Z., Zhang, H., Chen, X., Zhang, C., Ma, W., Huang, C., ... Dou, Z. (2018). Pursuing sustainable productivity with millions of smallholder farmers. *Nature*, 555(7696), 363–366. <https://doi.org/10.1038/nature25785>
- Curie, C., Cassin, G., Couch, D., Divol, F., Higuchi, K., Le Jean, M., ... Mari, S. (2009). Metal movement within the plant: Contribution of nicotianamine and yellow stripe 1-like transporters. *Annals of Botany*, 103(1), 1–11. <https://doi.org/10.1093/aob/mcn207>
- Curtis, T., & Halford, N. G. (2014). Food security: The challenge of increasing wheat yield and the importance of not compromising food safety. *Annals of Applied Biology*, 164(3), 354–372. <https://doi.org/10.1111/aab.12108>
- Das, S., & Green, A. (2013). Importance of zinc in crops and human health. *Journal of SAT Agricultural Research*, 11, 1–7.
- Davis, D. R. (2009). Declining fruit and vegetable nutrient composition: What is the evidence? *HortScience*, 44(1), 15–19. <https://doi.org/10.21273/HORTSCI.44.1.15>
- de Onis, M., Monteiro, C., Akre, J., & Clugston, G. (1993). The worldwide magnitude of protein-energy malnutrition: An overview from the WHO Global Database on Child Growth. *Bulletin of the World Health Organization*, 71(6), 703–712.
- Domnguez, E., Heredia-Guerrero, J., & Heredia, A. (2011). The biophysical design of plant cuticles: An overview. *New Phytologist*, 189(4), 938–949. <https://doi.org/10.1111/j.1469-8137.2010.03553.x>

- Durmaz, E., Coruh, C., Dinler, G., Grusak, M. A., Peleg, Z., Saranga, Y., ... Budak, H. (2011). Expression and cellular localization of ZIP1 transporter under zinc deficiency in wild emmer wheat. *Plant Molecular Biology Reporter*, 29(3), 582–596. <https://doi.org/10.1007/s11105-010-0264-3>
- Eagling, T., Neal, A. L., McGrath, S. P., Fairweather-Tait, S., Shewry, P. R., & Zhao, F.-J. (2014). Distribution and speciation of iron and zinc in grain of two wheat genotypes. *Journal of Agricultural and Food Chemistry*, 62(3), 708–716. <https://doi.org/10.1021/jf403331p>
- Erenoglu, B., Nikolic, M., Römhelt, V., & Cakmak, I. (2002). Uptake and transport of foliar applied zinc (^{65}Zn) in bread and durum wheat cultivars differing in zinc efficiency. *Plant and Soil*, 241(2), 251–257. <https://doi.org/10.1023/A:1016148925918>
- Erenoglu, E. B., Kutman, U. B., Ceylan, Y., Yildiz, B., & Cakmak, I. (2011). Improved nitrogen nutrition enhances root uptake, root-to-shoot translocation and remobilization of zinc (^{65}Zn) in wheat. *New Phytologist*, 189(2), 438–448. <https://doi.org/10.1111/j.1469-8137.2010.03488.x>
- Evens, N. P., Buchner, P., & Williams, L. E. (2017). The role of ZIP transporters and group F bZIP transcription factors in the Zn-deficiency response of wheat (*Triticum aestivum*). *Plant Journal*, 92(2), 291–304. <https://doi.org/10.1111/tpj.13655>
- Fan, M. S., Zhao, F. J., Fairweather-Tait, S. J., Poulton, P. R., Sunham, S. J., & McGrath, S. P. (2008). Evidence of decreasing mineral density in wheat grain over the last 160 years. *Journal of Trace Elements in Medicine and Biology*, 22(4), 315–324. <https://doi.org/10.1016/j.jtemb.2008.07.002>
- Feng, G., Xu, Q., Wang, Z., & Zhuoma, Q. (2016). AINTEGUMENTA negatively regulates age-dependent leaf senescence downstream of AUXIN RESPONSE FACTOR 2 in *Arabidopsis thaliana*. *Plant Biotechnology*, 33(2), 71–76. <https://doi.org/10.5511/plantbiotechnology.16.0222a>
- Fernández, V., Bahamonde, H. A., Javier Peguero-Pina, J., Gil-Pelegrín, E., Sancho-Knapik, D., Gil, L., ... Eichert, T. (2017). Physico-chemical properties of plant cuticles and their functional and ecological significance. *Journal of Experimental Botany*, 68(19), 5293–5306. <https://doi.org/10.1093/jxb/erx302>
- Fernández, V., & Brown, P. H. (2013). From plant surface to plant metabolism: The uncertain fate of foliar-applied nutrients. *Frontiers in Plant Science*, 4, 289. <https://doi.org/10.3389/fpls.2013.00289>
- Foulkes, M. J., Slafer, G. A., Davies, W. J., Berry, P. M., Sylvester-Bradley, R., Martre, P., ... Reynolds, M. P. (2011). Raising yield potential of wheat. III. Optimizing partitioning to grain while maintaining lodging resistance. *Journal of Experimental Botany*, 62(2), 469–486. <https://doi.org/10.1093/jxb/erq300>
- Foy, C. D., Montenegro, G., & Barber, S. A. (1953). Foliar feeding of corn with urea nitrogen. *Soil Science Society of America Journal*, 17(4), 387–390. <https://doi.org/10.2136/sssaj1953.03615995001700040022x>
- Gao, X., Mohr, R. M., McLaren, D. L., & Grant, C. A. (2011). Grain cadmium and zinc concentrations in wheat as affected by genotypic variation and potassium chloride fertilization. *Field Crops Research*, 122(2), 95–103. <https://doi.org/10.1016/j.fcr.2011.03.005>
- Garnett, T. P., & Graham, R. D. (2005). Distribution and remobilization of iron and copper in wheat. *Annals of Botany*, 95(5), 817–826. <https://doi.org/10.1093/aob/mci085>
- Garvin, D. F., Welch, R. M., & Finley, J. W. (2006). Historical shifts in the seed mineral micronutrient concentrations of US hard red winter wheat germplasm. *Journal of the Science of Food and Agriculture*, 86(13), 2213–2220. <https://doi.org/10.1002/jsfa.2601>
- Gemenet, D., Hash, C., Sanogo, M., Sy, O., Zangre, R., Leiser, W., & Haussmann, B. (2015). Phosphorus uptake and utilization efficiency in West African pearl millet inbred lines. *Field Crops Research*, 171, 54–66. <https://doi.org/10.1016/j.fcr.2014.11.001>
- Gibson, R. S., Bailey, K. B., Gibbs, M., & Ferguson, E. L. (2010). A review of phytate, iron, zinc and calcium concentrations in plant-based complementary foods used in low-income countries and implications for bioavailability. *Food and Nutrition Bulletin*, 31(2_suppl2), S134–S146. <https://doi.org/10.1177/15648265100312S206>
- Gomez-Coronado, F., Poblaciones, M. J., Almeida, A. S., & Cakmak, I. (2016). Zinc (Zn) concentration of bread wheat grown under Mediterranean conditions as affected by genotype and soil/foliar Zn application. *Plant and Soil*, 401(1–2), 331–346. <https://doi.org/10.1007/s11104-015-2758-0>
- Gooding, M., Fan, M., McGrath, S., Shewry, P., & Zhao, F. (2012). Contrasting effects of dwarfing alleles and nitrogen availability on mineral concentrations in wheat grain. *Plant and Soil*, 360(1–2), 93–107. <https://doi.org/10.1007/s11104-012-1203-x>
- Graham, R. D., Welch, R. M., Saunders, D. A., Ortiz-Monasterio, I., Bouis, H. E., Bonierbale, M., ... Twomlow, S. (2007). Nutritious subsistence food systems. *Advances in Agronomy*, 92, 1–74. [https://doi.org/10.1016/S0065-2113\(04\)92001-9](https://doi.org/10.1016/S0065-2113(04)92001-9)
- Grassini, P., Eskridge, K. M., & Cassman, K. G. (2013). Distinguishing between yield advances and yield plateaus in historical crop production trends. *Nature Communications*, 4, 2918. <https://doi.org/10.1038/ncomms3918>
- Grotz, N., Fox, T., Connolly, E., Park, W., Gueriot, M. L., & Eide, D. (1998). Identification of a family of zinc transporter genes from *Arabidopsis* that respond to zinc deficiency. *Proceedings of the National Academy of Sciences of the United States of America*, 95(12), 7220–7224. <https://doi.org/10.1073/pnas.95.12.7220>
- Grusak, M. A., Pearson, J. N., & Marentes, E. (1999). The physiology of micronutrient homeostasis in field crops. *Field Crops Research*, 60(1–2), 41–56. [https://doi.org/10.1016/S0378-4290\(98\)00132-4](https://doi.org/10.1016/S0378-4290(98)00132-4)
- Gueriot, M. L. (2000). The ZIP family of metal transporters. *Biochimica Et Biophysica Acta (BBA) – Biomembranes*, 1465(1–2), 190–198. [https://doi.org/10.1016/S0005-2736\(00\)00138-3](https://doi.org/10.1016/S0005-2736(00)00138-3)
- Gupta, N., Ram, H., & Kumar, B. (2016). Mechanism of zinc absorption in plants: Uptake, transport, translocation and accumulation. *Reviews in Environmental Science and Bio/Technology*, 15, 89–109. <https://doi.org/10.1007/s11157-016-9390-1>
- Gupta, O. P., Pandey, V., Saini, R., Narwal, S., Malik, V. K., Khandale, T., ... Singh, G. P. (2020). Identifying transcripts associated with efficient transport and accumulation of Fe and Zn in hexaploid wheat (*T. aestivum* L.). *Journal of Biotechnology*, 316, 46–55. <https://doi.org/10.1016/j.jbiotec.2020.03.015>
- Hao, M.-D., Wei, X.-R., & Dang, T.-H. (2003). Effect of long-term applying zinc fertilizer on wheat yield and content of zinc in dryland. *Journal of Plant Nutrition and Fertilizers*, 9(3), 377–380 (in Chinese). <https://doi.org/10.11674/zwyf.2003.0324>
- Hao, Y. F., Zhang, Y., & He, Z. H. (2015). Progress in zinc biofortification of crops. *Chinese Bulletin of Life Sciences*, 27(8), 1047–1054. <https://doi.org/10.13376/j.cbbs/2015144>
- Haslett, B. S., Reid, R. J., & Rengel, Z. (2001). Zinc mobility in wheat: Uptake and distribution of zinc applied to leaves or roots. *Annals of Botany*, 87(3), 379–386. <https://doi.org/10.1006/anbo.2000.1349>

- Hoffland, E., Wei, C., & Wissuwa, M. (2006). Organic anion exudation by lowland rice (*Oryza sativa* L.) at zinc and phosphorus deficiency. *Plant and Soil*, 283(1–2), 155–162. <https://doi.org/10.1007/s11104-005-3937-1>
- Hotz, C., & Brown, K. H. (2004). Assessment of the risk of zinc deficiency in populations and options for its control. *Food and Nutrition Bulletin*, 25(1_suppl2), S94–S203. <https://doi.org/10.4067/S0717-75182010000200014>
- Hussain, D., Haydon, M. J., Wang, Y., Wong, E., Sherson, S. M., Young, J., ... Cobbett, C. S. (2004). P-type ATPase heavy metal transporters with roles in essential zinc homeostasis in *Arabidopsis*. *The Plant Cell*, 16(5), 1327–1339. <https://doi.org/10.1105/tpc.020487>
- Ishimaru, Y., Suzuki, M., Kobayashi, T., Takahashi, M., Nakanishi, H., Mori, S., & Nishizawa, N. K. (2005). OsZIP4, a novel zinc-regulated zinc transporter in rice. *Journal of Experimental Botany*, 56(422), 3207–3214. <https://doi.org/10.1093/jxb/eri317>
- Karim, M., Zhang, Y., Zhao, R., Chen, X., Zhang, F., & Zou, C. (2012). Alleviation of drought stress in winter wheat by late foliar application of zinc, boron, and manganese. *Journal of Plant Nutrition and Soil Science*, 175(1), 142–151. <https://doi.org/10.1002/jpln.2011010141>
- Kim, J. I., Murphy, A. S., Baek, D., Lee, S.-W., Yun, D.-J., Bressan, R. A., & Narasimhan, M. L. (2011). *YUCCA6* over-expression demonstrates auxin function in delaying leaf senescence in *Arabidopsis thaliana*. *Journal of Experimental Botany*, 62(11), 3981–3992. <https://doi.org/10.1093/jxb/err094>
- Krežel, A., & Maret, W. (2016). The biological inorganic chemistry of zinc ions. *Archives of Biochemistry and Biophysics*, 611, 3–19. <https://doi.org/10.1016/j.abb.2016.04.010>
- Kutman, U., Kutman, B., Ceylan, Y., Ova, E., & Cakmak, I. (2012). Contributions of root uptake and remobilization to grain zinc accumulation in wheat depending on post-anthesis zinc availability and nitrogen nutrition. *Plant and Soil*, 361(1–2), 177–187. <https://doi.org/10.1007/s11104-012-1300-x>
- Kutman, U., Yildiz, B., & Cakmak, I. (2011). Effect of nitrogen on uptake, remobilization and partitioning of zinc and iron throughout the development of durum wheat. *Plant and Soil*, 342(1–2), 149–164. <https://doi.org/10.1007/s11104-010-0679-5>
- Kutman, U. B., Yildiz, B., Ozturk, L., & Cakmak, I. (2010). Biofortification of durum wheat with zinc through soil and foliar applications of nitrogen. *Cereal Chemistry*, 87(1), 1–9. <https://doi.org/10.1094/CCHEM-87-1-0001>
- Leakey, A. D. B. (2009). Rising atmospheric carbon dioxide concentration and the future of C₄ crops for food and fuel. *Proceedings of the Royal Society B: Biological Sciences*, 276(1666), 2333–2343. <https://doi.org/10.1098/rspb.2008.1517>
- Lee, I. C., Hong, S. W., Whang, S. S., Lim, P. O., Nam, H. G., & Koo, J. C. (2011). Age-dependent action of an ABA-inducible receptor kinase, RPK1, as a positive regulator of senescence in *Arabidopsis* leaves. *Plant and Cell Physiology*, 52(4), 651–662. <https://doi.org/10.1093/pcp/pcr026>
- Li, H., Huang, G., Meng, Q., Ma, L., Yuan, L., Wang, F., ... Zhang, F. (2011). Integrated soil and plant phosphorus management for crop and environment in China. A review. *Plant and Soil*, 349(1–2), 157–167. <https://doi.org/10.1007/s11104-011-0909-5>
- Li, S., Zhou, X., Huang, Y., Zhu, L., Zhang, S., Zhao, Y., ... Chen, R. (2013). Identification and characterization of the zinc-regulated transporters, iron-regulated transporter-like protein (ZIP) gene family in maize. *BMC Plant Biology*, 13, 114. <https://doi.org/10.1186/1471-2229-13-114>
- Li, T., Zhang, W., Yin, J., Chadwick, D., Norse, D., Lu, Y., ... Dou, Z. (2018). Enhanced-efficiency fertilizers are not a panacea for resolving the nitrogen problem. *Global Change Biology*, 24(2), e511–e521. <https://doi.org/10.1111/gcb.13918>
- Li, X., Kristiansen, K., Rosenqvist, E., & Liu, F. (2019). Elevated CO₂ modulates the effects of drought and heat stress on plant water relations and grain yield in wheat. *Journal of Agronomy and Crop Science*, 205(4), 362–371. <https://doi.org/10.1111/jac.12330>
- Li, X., Li, Y., Zhu, X., Liu, S., & Liu, F. (2019). Modulation of photosynthate supply by CO₂ elevation affects the post-head-emergence frost-induced grain yield loss in wheat. *Journal of Agronomy and Crop Science*, 205(1), 54–64. <https://doi.org/10.1111/jac.12304>
- Li, X., Ulfat, A., Lv, Z., Fang, L., Jiang, D., & Liu, F. (2019). Effect of multigenerational exposure to elevated atmospheric CO₂ concentration on grain quality in wheat. *Environmental and Experimental Botany*, 157, 310–319. <https://doi.org/10.1016/j.envexpbot.2018.10.028>
- Lim, P. O., Kim, H. J., & Nam, H. G. (2007). Leaf senescence. *Annual Review of Plant Biology*, 58, 115–136. <https://doi.org/10.1146/annurev.arplant.57.032905.105316>
- Lim, P. O., Lee, I. C., Kim, J., Kim, H. J., Ryu, J. S., Woo, H. R., & Nam, H. G. (2010). Auxin response factor 2 (ARF2) plays a major role in regulating auxin-mediated leaf longevity. *Journal of Experimental Botany*, 61(5), 1419–1430. <https://doi.org/10.1093/jxb/erq010>
- Liu, D., Liu, Y., Zhang, W., Chen, X., & Zou, C. (2017). Agronomic approach of zinc biofortification can increase zinc bioavailability in wheat flour and thereby reduce zinc deficiency in humans. *Nutrients*, 9(5), 465. <https://doi.org/10.3390/nu9050465>
- Liu, D.-Y., Liu, Y.-M., Zhang, W., Chen, X.-P., & Zou, C.-Q. (2019). Zinc uptake, translocation, and remobilization in winter wheat as affected by soil application of Zn fertilizer. *Frontiers in Plant Science*, 10, 426. <https://doi.org/10.3389/fpls.2019.00426>
- Liu, D., Zhang, W., Liu, Y., Chen, X., & Zou, C. (2020). Soil application of zinc fertilizer increases maize yield by enhancing the kerner number and kernel weight of inferior grains. *Frontiers in Plant Science*, 11, 188. <https://doi.org/10.3389/fpls.2020.00188>
- Liu, N., Zhang, Y., Wang, B., Xue, Y., Yu, P., Zhang, Q., & Wang, Z. (2014). Is grain zinc concentration in wheat limited by source? *Australian Journal of Crop Science*, 8, 1534–1541.
- Liu, Y.-M., Liu, D.-Y., Zhang, W., Chen, X.-X., Zhao, Q.-Y., Chen, X.-P., & Zou, C.-Q. (2020). Health risk assessment of heavy metals (Zn, Cu, Cd, Pb, As and Cr) in wheat grain receiving repeated Zn fertilizers. *Environmental Pollution*, 257, 113581. <https://doi.org/10.1016/j.envpol.2019.113581>
- Loneragan, J. F., Grove, T. S., Robson, A. D., & Snowball, K. (1979). Phosphorus toxicity as a factor in zinc-phosphorus interactions in plants. *Soil Science Society of America Journal*, 43(5), 966–972. <https://doi.org/10.2136/sssaj1979.03615995004300050031x>
- Lv, X., Zhang, Y., Zhang, Y., Fan, S., & Kong, L. (2019). Source-sink modifications affect leaf senescence and grain mass in wheat. *bioRxiv*, 647743. <https://doi.org/10.1101/647743>
- Ma, G. S., Jin, Y., Piao, J., Kok, F., Guusje, B., & Jacobsen, E. (2005). Phytate, calcium, iron, and zinc contents and their molar ratios in foods commonly consumed in China. *Journal of Agricultural and Food Chemistry*, 53(26), 10285–10290. <https://doi.org/10.1021/jf052051r>
- Ma, Y.-Z., MacKown, C. T., & Van Sanford, D. A. (1996). Differential effects of partial spikelet removal and defoliation on kernel growth and assimilate partitioning among wheat cultivars. *Field Crops Research*, 47(2–3), 201–209. [https://doi.org/10.1016/0378-4290\(96\)00016-0](https://doi.org/10.1016/0378-4290(96)00016-0)

- MacDonald, G. K., Bennett, E. M., Potter, P. A., & Ramankutty, N. (2011). Agronomic phosphorus imbalances across the world's croplands. *Proceedings of the National Academy of Sciences of the United States of America*, 108(7), 3086–3091. <https://doi.org/10.1073/pnas.1010808108>
- Mandal, B., & Mandal, L. N. (1990). Effect of phosphorus application on transformation of zinc fraction in soil and on the zinc nutrition of lowland rice. *Plant and Soil*, 121(1), 115–123. <https://doi.org/10.1007/bf00013104>
- Marschner, P. (2012). *Marschner's mineral nutrition of higher plants* (3rd ed.). San Diego, CA: Academic Press, Elsevier.
- Mason, T., & Maskell, E. (1928). Studies of the transport of carbohydrate in the cotton plant: II. The factors determining the rate and the direction of movement of sugars. *Annals of Botany*, 42(3), 571–636. <https://doi.org/10.1093/oxfordjournals.aob.a090131>
- Mengel, K., & Kirkby, E. (1987). *Principles of plant nutrition*. Bern, Germany: Lang Druck, Liebefeld.
- Milani, N., McLaughlin, M. J., Stacey, S. P., Kirby, J. K., Hettiarachchi, G. M., Beak, D. G., & Cornelis, G. (2012). Dissolution kinetics of macronutrient fertilizers coated with manufactured zinc oxide nanoparticles. *Journal of Agricultural and Food Chemistry*, 60(16), 3991–3998. <https://doi.org/10.1021/jf205191y>
- Milner, M., Seamon, J., Craft, E., & Kochian, L. (2013). Transport properties of members of the ZIP family in plants and their role in Zn and Mn homeostasis. *Journal of Experimental Botany*, 64(1), 369–381. <https://doi.org/10.1093/jxb/ers315>
- Mohammad, F., & Naseem, U. (2006). Effect of K application on leaf carbonic anhydrase and nitrate reductase activities, photosynthetic characteristics, NPK and NO₃ contents, growth, and yield of mustard. *Photosynthetica*, 44(3), 471–473. <https://doi.org/10.1007/s11099-006-0053-2>
- Moreno-Jiménez, E., Plaza, C., Saiz, H., Manzano, R., Flagmeier, M., & Maestre, F. T. (2019). Aridity and reduced soil micronutrient availability in global drylands. *Nature Sustainability*, 2, 371–377. <https://doi.org/10.1038/s41893-019-0262-x>
- Morgounov, A., Gómez-Becerra, H. F., Abugalieva, A., Dzhunusova, M., Yessimbekova, M., Muminjanov, H., ... Cakmak, I. (2007). Iron and zinc grain density in common wheat grown in Central Asia. *Euphytica*, 155, 193–203. <https://doi.org/10.1007/s10668-006-9321-2>
- Myers, S. S., Zanolletti, A., Kloog, I., Huybers, P., Leakey, A. D. B., Bloom, A. J., ... Usui, Y. (2014). Increasing CO₂ threatens human nutrition. *Nature*, 510, 139–142. <https://doi.org/10.1038/nature13179>
- Nawaz, F., Ahmad, R., Ashraf, M. Y., Waraich, E. A., & Khan, S. Z. (2015). Effect of selenium foliar spray on physiological and biochemical processes and chemical constituents of wheat under drought stress. *Ecotoxicology and Environmental Safety*, 113, 191–200. <https://doi.org/10.1016/j.ecoenv.2014.12.003>
- Nevo, Y., & Nelson, N. (2006). The NRAMP family of metal-ion transporters. *Biochimica Et Biophysica Acta (BBA) – Molecular Cell Research*, 1763(7), 609–620. <https://doi.org/10.1016/j.bbamcr.2006.05.007>
- Nie, Z., Zhao, P., Shi, H., Wang, Y., Qin, S., & Liu, H. (2019). Nitrogen supply enhances zinc uptake and root-to-shoot translocation via up-regulating the expression of *TaZIP3* and *TaZIP7* in winter wheat (*Triticum aestivum*). *Plant and Soil*, 444(1–2), 501–517. <https://doi.org/10.1007/s11104-019-04295-4>
- Nowack, B., Schwyzer, I., & Schulin, R. (2008). Uptake of Zn and Fe by wheat (*Triticum aestivum* var. Greina) and transfer to the grain in the presence of chelating agents (Ethylenediaminedisuccinic acid and Ethylenediaminetetraacetic acid). *Journal of Agricultural and Food Chemistry*, 56(12), 4643–4649. <https://doi.org/10.1021/jf800041b>
- Ortiz-Monasterio, I., Cardenas, M. E., & Cakmak, I. (2015). Zinc bio-fortification in wheat through foliar fertilization combined with pesticides. Paper presented at the 4th International Zinc Symposium: Improving Crop Production and Human Health, Sao Paulo.
- Ortiz-Monasterio, J. I., Palacios-Rojas, N., Meng, E., Pixley, K., Trethowan, R., & Peña, R. J. (2007). Enhancing the mineral and vitamin content of wheat and maize through plant breeding. *Journal of Cereal Science*, 46(3), 293–297. <https://doi.org/10.1016/j.jcs.2007.06.005>
- Ota, E., Mori, R., Middleton, P., Tobe-Gai, R., Mahomed, K., Miyazaki, C., & Bhutta, Z. A. (2015). Zinc supplementation for improving pregnancy and infant outcome. *Cochrane Database of Systematic Reviews*, 2, CD000230. <https://doi.org/10.1002/14651858.CD000230.pub5>
- Ova, E., Kutman, U., Ozturk, L., & Cakmak, I. (2015). High phosphorus supply reduced zinc concentration of wheat in native soil but not in autoclaved soil or nutrient solution. *Plant and Soil*, 393(1–2), 147–162. <https://doi.org/10.1007/s11104-015-2483-8>
- Ozturk, L., Yazici, M. A., Yucel, C., Torun, A., Cekic, C., Bagci, A., ... Cakmak, I. (2006). Concentration and localization of zinc during seed development and germination in wheat. *Physiologia Plantarum*, 128(1), 144–152. <https://doi.org/10.1111/j.1399-3054.2006.00737.x>
- Palmer, C. M., & Gueriot, M. L. (2009). Facing the challenges of Cu, Fe and Zn homeostasis in plants. *Nature Chemical Biology*, 5(5), 333–340. <https://doi.org/10.1038/nchembio.166>
- Palmer, L. J., Dias, D. A., Boughton, B., Roessner, U., Graham, R. D., & Stangoulis, J. C. R. (2014). Metabolite profiling of wheat (*Triticum aestivum* L.) phloem exudate. *Plant Methods*, 10, 27. <https://doi.org/10.1186/1746-4811-10-27>
- Pearce, S., Tabbita, F., Cantu, D., Buffalo, V., Avni, R., Vazquez-Gross, H., ... Dubcovsky, J. (2014). Regulation of Zn and Fe transporters by the *GPC1* gene during early wheat monocarpic senescence. *BMC Plant Biology*, 14, 368. <https://doi.org/10.1186/s12870-014-0368-2>
- Pearson, J. N., Jenner, C. F., Rengel, Z., & Graham, R. D. (1996). Differential transport of Zn, Mn and sucrose along the longitudinal axis of developing wheat grains. *Physiologia Plantarum*, 97(2), 332–338. <https://doi.org/10.1034/j.1399-3054.1996.970217.x>
- Pearson, J. N., Rengel, Z., Jenner, C. F., & Graham, R. D. (1996). Manipulation of xylem transport affects Zn and Mn transport into developing wheat grains of cultured ears. *Physiologia Plantarum*, 98(2), 229–234. <https://doi.org/10.1034/j.1399-3054.1996.980202.x>
- Persson, D. P., Bang, T. C., Pedas, P. R., Kutman, U. B., Cakmak, I., Andersen, B., ... Husted, S. (2016). Molecular speciation and tissue compartmentation of zinc in durum wheat grains with contrasting nutritional status. *New Phytologist*, 211(4), 1255–1265. <https://doi.org/10.1111/nph.13989>
- Pfeiffer, W. H., & McClafferty, B. (2007). Biofortification: Breeding micronutrient-dense crops. In M. S. Kang, & P. M. Priyadarshan (Eds.), *Breeding major food staples* (pp. 61–91). Oxford, UK: Blackwell Publishing.
- Poursarebani, N., Nussbaumer, T., Šimková, H., Šafář, J., Witsenboer, H., van Oeveren, J., ... Schnurbusch, T. (2014). Whole-genome profiling and shotgun sequencing delivers an anchored, gene-decorated, physical map assembly of bread wheat chromosome 6A. *The Plant Journal*, 79, 334–347. <https://doi.org/10.1111/tbj.12550>

- Prasad, A. S. (2013). Discovery of human zinc deficiency: Its impact on human health and disease. *Advances in Nutrition*, 4(2), 176–190. <https://doi.org/10.3945/an.112.003210>
- Rakshit, R., Patra, A. K., Purakayastha, T. J., Singh, R. D., Pathak, H., & Dhar, S. (2016). Super-optimal NPK along with foliar iron application influences bioavailability of iron and zinc of wheat. *Proceedings of the National Academy of Sciences, India Section B: Biological Sciences*, 86, 159–164. <https://doi.org/10.1007/s40011-014-0428-2>
- Ram, H., Rashid, A., Zhang, W., Duarte, A. P., Phattarakul, N., Simunji, S., ... Cakmak, I. (2016). Biofortification of wheat, rice and common bean by applying foliar zinc fertilizer along with pesticides in seven countries. *Plant and Soil*, 403, 389–401. <https://doi.org/10.1007/s11104-016-2815-3>
- Rengel, Z. (2015). Availability of Mn, Zn and Fe in the rhizosphere. *Journal of Soil Science and Plant Nutrition*, 15(2), 397–409.
- Ricachenevsky, F., Menguer, P., Sperotto, R., Williams, L., & Fett, J. (2013). Roles of plant metal tolerance proteins (MTP) in metal storage and potential use in biofortification strategies. *Frontiers in Plant Science*, 4, 144. <https://doi.org/10.3389/fpls.2013.00144>
- Rodgers, A., Ezzati, M., Hoorn, S. V., Lopez, A. D., Lin, R.-B., & Murray, C. J. L. (2004). Distribution of major health risks: Findings from the global burden of disease study. *PLoS Med*, 1(1), e27. <https://doi.org/10.1371/journal.pmed.0010027>
- Roelfsema, M. R. G., & Hedrich, R. (2005). In the light of stomatal opening: New insights into 'the Watergate'. *New Phytologist*, 167(3), 665–691. <https://doi.org/10.1111/j.1469-8137.2005.01460.x>
- Rose, T., & Bowden, B. (2013). Matching soil nutrient supply and crop demand during the growing Season. In Z. Rengel (Ed.), *Improving water- and nutrient-use efficiency in food production systems* (pp. 93–103). Chichester UK: Wiley-Blackwell.
- Sah, S., Reddy, K., & Li, J. (2016). Absciscic acid and abiotic stress tolerance in crop plants. *Frontiers in Plant Science*, 7, 571. <https://doi.org/10.3389/fpls.2016.00571>
- Sasaki, H., Edo, E. I., Uehara, N., Ishimaru, T., Kawamitsu, Y., Suganuma, S., ... Ohsugi, R. (2005). Effect of sucrose on activity of starch synthesis enzymes in rice ears in culture. *Physiologia Plantarum*, 124(3), 301–310. <https://doi.org/10.1111/j.1399-3054.2005.00520.x>
- Schreiber, L. (2005). Polar paths of diffusion across plant cuticles: New evidence for an old hypothesis. *Annals of Botany*, 95(7), 1069–1073. <https://doi.org/10.1093/aob/mci122>
- Sharma, P., Aggarwal, P., & Kaur, A. (2017). Biofortification: A new approach to eradicate hidden hunger. *Food Reviews International*, 33(1), 1–21. <https://doi.org/10.1080/87559129.2015.1137309>
- Shewry, P. R., Pellny, T. K., & Lovegrove, A. (2016). Is modern wheat bad for health? *Nature Plants*, 2(7), 1–3. <https://doi.org/10.1038/nplants.2016.97>
- Shi, R., Zhang, Y., Chen, X., Sun, Q., Zhang, F., Römhelt, V., & Zou, C. (2010). Influence of long-term nitrogen fertilization on micronutrient density in grain of winter wheat (*Triticum aestivum* L.). *Journal of Cereal Science*, 51(1), 165–170. <https://doi.org/10.1016/j.jcs.2009.11.008>
- Singh, B. R., Timsina, Y. N., Lind, O. C., Cagno, S., & Janssens, K. (2018). Zinc and iron concentration as affected by nitrogen fertilization and their localization in wheat grain. *Frontiers in Plant Science*, 9, 307. <https://doi.org/10.3389/fpls.2018.00307>
- Singh, D., & Prasanna, R. (2020). Potential of microbes in the biofortification of Zn and Fe in dietary food grains. A review. *Agronomy for Sustainable Development*, 40, 15. <https://doi.org/10.1007/s13593-020-00619-2>
- Sperotto, R. A. (2013). Zn/Fe remobilization from vegetative tissues to rice seeds: Should I stay or should I go? Ask Zn/Fe supply!. *Frontiers in Plant Science*, 4, 464. <https://doi.org/10.3389/fpls.2013.00464>
- Srivastava, P. C., Ansari, U. I., Pachauri, S. P., & Tyagi, A. K. (2016). Effect of zinc application methods on apparent utilization efficiency of zinc and potassium fertilizers under rice-wheat rotation. *Journal of Plant Nutrition*, 39(3), 348–364. <https://doi.org/10.1080/01904167.2015.1016170>
- Stein, A. J. (2010). Global impacts of human mineral malnutrition. *Plant and Soil*, 335(1–2), 133–154. <https://doi.org/10.1007/s11104-009-0228-2>
- Tang, J. W., Zou, C. Q., He, Z. H., Shi, R. L., Ortiz-Monasterio, I., Qu, Y. Y., & Zhang, Y. (2008). Mineral element distributions in milling fractions of Chinese wheats. *Journal of Cereal Science*, 48(3), 821–828. <https://doi.org/10.1016/j.jcs.2008.06.008>
- Tao, Z.-Q., Wang, D.-M., Chang, X.-H., Wang, Y.-J., Yang, Y.-S., & Zhao, G.-C. (2018). Effects of zinc fertilizer and short-term high temperature stress on wheat grain production and wheat flour proteins. *Journal of Integrative Agriculture*, 17(9), 1979–1990. [https://doi.org/10.1016/S2095-3119\(18\)61911-2](https://doi.org/10.1016/S2095-3119(18)61911-2)
- Teng, W., Deng, Y., Chen, X.-P., Xu, X.-F., Chen, R.-Y., Lv, Y., ... Li, Z.-S. (2013). Characterization of root response to phosphorus supply from morphology to gene analysis in field-grown wheat. *Journal of Experimental Botany*, 64(5), 1403–1411. <https://doi.org/10.1093/jxb/ert023>
- Tilman, D., Cassman, K. G., Matson, P. A., Naylor, R., & Polasky, S. (2002). Agricultural sustainability and intensive production practices. *Nature*, 418(6898), 671–677. <https://doi.org/10.1038/nature01014>
- Tiong, J., McDonald, G. K., Genc, Y., Pedas, P., Hayes, J. E., Toubia, J., ... Huang, C. Y. (2014). HvZIP7 mediates zinc accumulation in barley (*Hordeum vulgare*) at moderately high zinc supply. *New Phytologist*, 201(1), 131–143. <https://doi.org/10.1111/nph.12468>
- Tiong, J., McDonald, G., Genc, Y., Shirley, N., Langridge, P., & Huang, C. Y. (2015). Increased expression of six ZIP family genes by zinc (Zn) deficiency is associated with enhanced uptake and root-to-shoot translocation of Zn in barley (*Hordeum vulgare*). *New Phytologist*, 207(4), 1097–1109. <https://doi.org/10.1111/nph.13413>
- Uauy, C., Distelfeld, A., Fahima, T., Blechl, A., & Dubcovsky, J. (2006). A NAC gene regulating senescence improves grain protein, zinc, and iron content in wheat. *Science*, 314(5803), 1298–1301. <https://doi.org/10.1126/science.1133649>
- United Nations International Children's Emergency Fund (2012). *Report-pneumonia and diarrhoea: Tackling the deadliest diseases for the world's poorest children* (p. 77). New York, NY: UNICEF.
- Velu, G., Ortiz-Monasterio, I., Cakmak, I., Hao, Y., & Singh, R. P. (2014). Biofortification strategies to increase grain zinc and iron concentrations in wheat. *Journal of Cereal Science*, 59(3), 365–372. <https://doi.org/10.1016/j.jcs.2013.09.001>
- Velu, G., Ortiz-Monasterio, I., Singh, R. P., & Payne, T. (2011). Variation for grain micronutrients concentration in wheat core-collection accessions of diverse origin. *Asian Journal of Crop Science*, 3, 43–48. <https://doi.org/10.3923/ajcs.2011.43.48>
- Velu, G., Singh, R. P., Huerta-Espino, J., Peña, R. J., Arun, B., Mahendru-Singh, A., ... Pfeiffer, W. H. (2012). Performance of biofortified spring wheat genotypes in target environments for grain zinc and iron concentrations. *Field Crops Research*, 137, 261–267. <https://doi.org/10.1016/j.fcr.2012.07.018>
- Vitousek, P. M., Naylor, R., Crews, T., David, M. B., Drinkwater, L. E., Holland, E., ... Zhang, F. S. (2009). Nutrient imbalances in

- agricultural development. *Science*, 324(5934), 1519–1520. <https://doi.org/10.1126/science.1170261>
- Wang, J., Mao, H., Zhao, H., Huang, D., & Wang, Z. (2012). Different increases in maize and wheat grain zinc concentrations caused by soil and foliar applications of zinc in Loess Plateau, China. *Field Crops Research*, 135, 89–96. <https://doi.org/10.1016/j.fcr.2012.07.010>
- Wang, S., Li, M., Tian, X., Chen, Y., Li, S., Liu, K., & Jia, Z. (2018). Effects of combined foliar application of Zn with N, P, or K on Zn accumulation, distribution and translocation in wheat. *Journal of Plant Nutrition and Fertilizers*, 24(2), 296–305 (in Chinese). <https://doi.org/10.11674/zwjy.17178>
- Wang, X.-Z., Liu, D.-Y., Zhang, W., Wang, C.-J., Cakmak, I., & Zou, C.-Q. (2015). An effective strategy to improve grain zinc concentration of winter wheat, aphids prevention and farmers' income. *Field Crops Research*, 184, 74–79. <https://doi.org/10.1016/j.fcr.2015.08.015>
- Wang, Y. X., Specht, A., & Horst, W. J. (2011). Stable isotope labelling and zinc distribution in grains studied by laser ablation ICP-MS in an ear culture system reveals zinc transport barriers during grain filling in wheat. *New Phytologist*, 189(2), 428–437. <https://doi.org/10.1111/j.1469-8137.2010.03489.x>
- Wang, Y.-H., Zou, C.-Q., Mirza, Z., Li, H., Zhang, Z.-Z., Li, D.-P., ... Zhang, Y.-Q. (2016). Cost of agronomic biofortification of wheat with zinc in China. *Agronomy for Sustainable Development*, 36(3), 44. <https://doi.org/10.1007/s13593-016-0382-x>
- Wang, Z., Li, H., Li, X., Xin, C., Si, J., Li, S., ... Wang, F. (2019). Nano-ZnO priming induces salt tolerance by promoting photosynthetic carbon assimilation in wheat. *Archives of Agronomy and Soil Science*, 66(9), 1259–1273. <https://doi.org/10.1080/03650340.2019.1663508>
- Wang, Z., Pan, F., Liu, Q., Yuan, L., Duan, Z., & Yin, X. (2018). Effect of zinc application at jointing stage on zinc bioavailability in wheat grain. *Soils*, 50(6), 1222–1228. <https://doi.org/10.13758/j.cnki.tr.2018.06.026>. (in Chinese).
- Waters, B. M., Uauy, C., Dubcovsky, J., & Grusak, M. A. (2009). Wheat (*Triticum aestivum*) NAM proteins regulate the translocation of iron, zinc, and nitrogen compounds from vegetative tissues to grain. *Journal of Experimental Botany*, 60(15), 4263–4274. <https://doi.org/10.1093/jxb/erp257>
- Wazzike, H. E., Yousfi, B. E., & Serghat, S. (2015). Contributions of three upper leaves of wheat, either healthy or inoculated by *Bipolaris sorokiniana*, to yield and yield components. *Australian Journal of Crop Science*, 9(7), 629–637.
- Welch, R. M., Graham, R. D., & Cakmak, I. (2013). *Linking agricultural production practices to improving human nutrition and health*. Paper presented at the ICN2 Second International Conference on Nutrition Preparatory Technical Meeting, Rome, Italy.
- White, P. J., & Broadley, M. R. (2009). Biofortification of crops with seven mineral elements often lacking in human diets – Iron, zinc, copper, calcium, magnesium, selenium and iodine. *New Phytologist*, 182(1), 49–84. <https://doi.org/10.1111/j.1469-8137.2008.02738.x>
- Wilkinson, S., & Davies, W. (2002). ABA-based chemical signalling: The co-ordination of responses to stress in plants. *Plant, Cell and Environment*, 25(2), 195–210. <https://doi.org/10.1046/j.0016-8025.2001.00824.x>
- Woo, H. R., Kim, H. J., Lim, P. O., & Nam, H. G. (2019). Leaf senescence: Systems and dynamics aspects. *Annual Review of Plant Biology*, 70, 347–376. <https://doi.org/10.1146/annurev-arplant-050718-095859>
- Wu, C., Dun, Y., Zhang, Z., Li, M., & Wu, G. (2020). Foliar application of selenium and zinc to alleviate wheat (*Triticum aestivum* L.) cadmium toxicity and uptake from cadmium-contaminated soil. *Ecotoxicology and Environmental Safety*, 190, 110091. <https://doi.org/10.1016/j.ecoenv.2019.110091>
- Wu, D., Dong, J., Yao, Y.-J., Zhao, W.-C., & Gao, X. (2015). Spatiotemporal expression pattern analysis of NAM transcription factors Gpc-1 and Gpc-2 in bread wheat cultivar Chinese Spring during grain filling. *Scientia Agricultura Sinica*, 48(7), 1262–1276 (in Chinese). <https://doi.org/10.3864/j.issn.0578-1752.2015.07.02>
- Xia, H., Kong, W., Wang, L., Xue, Y., Liu, W., Zhang, C., ... Li, C. (2019). Foliar Zn spraying simultaneously improved concentrations and bioavailability of Zn and Fe in maize grains irrespective of foliar sucrose supply. *Agronomy*, 9(7), 386. <https://doi.org/10.3390/agronomy9070386>
- Xia, H., Xue, Y., Kong, W., Tang, Y., Li, J., & Li, D. (2018). Effects of source/sink manipulation on grain zinc accumulation by winter wheat genotypes. *Chilean Journal of Agricultural Research*, 78(1), 117–125. <https://doi.org/10.4067/S0718-58392018000100117>
- Xia, H., Xue, Y., Liu, D., Kong, W., Xue, Y., Tang, Y., ... Mei, P. (2018). Rational application of fertilizer nitrogen to soil in combination with foliar Zn spraying improved Zn nutritional quality of wheat grains. *Frontiers in Plant Science*, 9, 677. <https://doi.org/10.3389/fpls.2018.00677>
- Xia, Q., Yang, Z. P., Xue, N. W., Dai, X. J., Zhang, X., & Gao, Z. Q. (2019). Effect of foliar application of selenium on nutrient concentration and yield of colored grain wheat in China. *Applied Ecology and Environmental Research*, 17(2), 2187–2202. https://doi.org/10.15666/aeer/1702_21872202
- Xue, Y.-F., Eagling, T., He, J., Zou, C.-Q., McGrath, S. P., Shewry, P. R., & Zhao, F.-J. (2014). Effects of nitrogen on the distribution and chemical speciation of iron and zinc in pearling fractions of wheat grain. *Journal of Agricultural and Food Chemistry*, 52(20), 4738–4746. <https://doi.org/10.1021/jf500273x>
- Xue, Y., Xia, H., Christie, P., Zhang, Z., Li, L., & Tang, C. (2016). Crop acquisition of phosphorus, iron and zinc from soil in cereal/legume intercropping systems: A critical review. *Annals of Botany*, 117(3), 363–377. <https://doi.org/10.1093/aob/mcv182>
- Xue, Y., Xia, H., McGrath, S., Shewry, P., & Zhao, F. (2015). Distribution of the stable isotopes ⁵⁷Fe and ⁶⁸Zn in grain tissues of various wheat lines differing in their phytate content. *Plant and Soil*, 396(1–2), 73–83. <https://doi.org/10.1007/s11104-015-2582-6>
- Xue, Y.-F., Yue, S.-C., Zhang, Y.-Q., Cui, Z.-L., Chen, X.-P., Yang, F.-C., ... Zou, C.-Q. (2012). Grain and shoot zinc accumulation in winter wheat affected by nitrogen management. *Plant and Soil*, 361(1–2), 153–163. <https://doi.org/10.1007/s11104-012-1510-2>
- Xue, Y., Zhang, W., Liu, D., Xia, H., & Zou, C. (2016). Nutritional composition of iron, zinc, calcium, and phosphorus in wheat grain milling fractions as affected by fertilizer nitrogen supply. *Cereal Chemistry*, 93(6), 543–549. <https://doi.org/10.1094/CCHEM-12-15-0243-R>
- Xue, Y.-F., Zhang, W., Liu, D.-Y., Yue, S.-C., Cui, Z.-L., Chen, X.-P., & Zou, C.-Q. (2014). Effects of nitrogen management on root morphology and zinc translocation from root to shoot of winter wheat in the field. *Field Crops Research*, 161, 38–45. <https://doi.org/10.1016/j.fcr.2014.01.009>
- Yang, J., & Zhang, J. (2018). Approach and mechanism in enhancing the remobilization of assimilates and grain-filling in rice and wheat. *Chinese Science Bulletin*, 63(28–29), 2932–2943. <https://doi.org/10.1360/N972018-00577>

- Yang, J., Zhang, J., Liu, K., Wang, Z., & Liu, L. (2006). Absciscic acid and ethylene interact in wheat grains in response to soil drying during grain filling. *New Phytologist*, 171(2), 293–303. <https://doi.org/10.1111/j.1469-8137.2006.01753.x>
- Yang, X. W., Tian, X. H., Lu, X. C., Cao, Y. X., & Chen, Z. H. (2011). Impacts of phosphorus and zinc levels on phosphorus and zinc nutrition and phytic acid concentration in wheat (*Triticum aestivum* L.). *Journal of the Science of Food and Agriculture*, 91(13), 2322–2328. <https://doi.org/10.1002/jsfa.4459>
- Zhang, M. (2017). *Investigations of the effects of Zn biofortification in different wheat genotypes*. PhD, Yangzhou University, Yangzhou, China.
- Zhang, W.-F., Dou, Z.-X., He, P., Ju, X.-T., Powlson, D., Chadwick, D., ... Zhang, F.-S. (2013). New technologies reduce greenhouse gas emissions from nitrogenous fertilizer in China. *Proceedings of the National Academy of Sciences of the United States of America*, 110(21), 8375–8380. <https://doi.org/10.1073/pnas.1210447110>
- Zhang, W., Liu, D., Li, C., Cui, Z., Chen, X., Russell, Y., & Zou, C. (2015). Zinc accumulation and remobilization in winter wheat as affected by phosphorus application. *Field Crops Research*, 184, 155–161. <https://doi.org/10.1016/j.fcr.2015.10.002>
- Zhang, W., Liu, D., Liu, Y., Chen, X., & Zou, C. (2017). Overuse of phosphorus fertilizer reduces the grain and flour protein contents and zinc bioavailability of winter wheat (*Triticum aestivum* L.). *Journal of Agricultural and Food Chemistry*, 65(8), 1473–1482. <https://doi.org/10.1021/acs.jafc.6b04778>
- Zhang, W., Liu, D., Liu, Y., Cui, Z., Chen, X., & Zou, C. (2016). Zinc uptake and accumulation in winter wheat relative to changes in root morphology and mycorrhizal colonization following varying phosphorus application on calcareous soil. *Field Crops Research*, 197, 74–82. <https://doi.org/10.1016/j.fcr.2016.08.010>
- Zhang, Y.-Q., Deng, Y., Chen, R.-Y., Cui, Z.-L., Chen, X.-P., Yost, R., ... Zou, C.-Q. (2012). The reduction in zinc concentration of wheat grain upon increased phosphorus-fertilization and its mitigation by foliar zinc application. *Plant and Soil*, 361(1–2), 143–152. <https://doi.org/10.1007/s11104-012-1238-z>
- Zhang, Y., Liu, G., Cheng, Y., Xu, J., Wang, C., & Yang, J. (2020). The effects of dry cultivation on grain-filling and chalky grains of upland rice and paddy rice. *Food and Energy Security*, 9(2), e198. <https://doi.org/10.1002/fes3.198>
- Zhang, Y. Q., Shi, R. L., Rezaul, K. M., Zhang, F. S., & Zou, C. Q. (2010). Iron and zinc concentrations in grain and flour of winter wheat as affected by foliar application. *Journal of Agricultural and Food Chemistry*, 58(23), 12268–12274. <https://doi.org/10.1021/jf103039k>
- Zhang, Y.-Q., Sun, Y.-X., Ye, Y.-L., Karim, M. R., Xue, Y.-F., Yan, P., ... Zou, C.-Q. (2012). Zinc biofortification of wheat through fertilizer applications in different locations of China. *Field Crops Research*, 125, 1–7. <https://doi.org/10.1016/j.fcr.2011.08.003>
- Zhang, Y., Zhang, Y., Liu, N., Su, D., Xue, Q., Stewart, B., & Wang, Z. (2012). Effect of source-sink manipulation on accumulation of micronutrients and protein in wheat grains. *Journal of Plant Nutrition and Soil Science*, 175(4), 622–629. <https://doi.org/10.1002/jpln.201100224>
- Zhang, Y.-H., Zhou, S.-L., Zhang, K., & Wang, Z.-M. (2008). Effects of source and sink reductions on micronutrient and protein contents of grain in wheat. *Acta Agronomica Sinica*, 34(9), 1629–1636. <https://doi.org/10.3724/SP.J.1006.2008.01629>
- Zhao, A. Q., Tian, X. H., Cao, Y. X., Lu, X. C., & Liu, T. (2014). Comparison of soil and foliar zinc application for enhancing grain zinc content of wheat when grown on potentially zinc-deficient calcareous soils. *Journal of the Science of Food and Agriculture*, 94(10), 2016–2022. <https://doi.org/10.1002/jsfa.6518>
- Zhao, A. Q., Wang, B. N., Tian, X. H., & Yang, X. B. (2020). Combined soil and foliar ZnSO₄ application improves wheat grain Zn concentration and Zn fractions in a calcareous soil. *European Journal of Soil Science*, 71(4), 681–694. <https://doi.org/10.1111/ejss.12903>
- Zou, C., Du, Y., Rashid, A., Ram, H., Savasli, E., Pieterse, P. J., ... Cakmak, I. (2019). Simultaneous biofortification of wheat with zinc, iodine, selenium, and iron through foliar treatment of a micronutrient cocktail in six countries. *Journal of Agricultural and Food Chemistry*, 2019(67), 8096–8106. <https://doi.org/10.1021/acs.jafc.9b01829>
- Zou, C., Du, Y., Rashid, A., Ram, H., Savasli, E., Pieterse, P. J., ... Cakmak, I. (2019). Simultaneous biofortification of wheat with zinc, iodine, selenium, and iron through foliar treatment of a micronutrient cocktail in six countries. *Journal of Agricultural and Food Chemistry*, 2019(67), 8096–8106. <https://doi.org/10.1021/acs.jafc.9b01829>
- Zou, C. Q., Zhang, Y. Q., Rashid, A., Ram, H., Savasli, E., Arisoy, R. Z., ... Cakmak, I. (2012). Biofortification of wheat with zinc through zinc fertilization in seven countries. *Plant and Soil*, 361(1–2), 119–130. <https://doi.org/10.1007/s11104-012-1369-2>
- Zuo, Z., Sun, L., Wang, T., Miao, P., Zhu, X., Liu, S., ... Li, X. (2017). Melatonin improves the photosynthetic carbon assimilation and antioxidant capacity in wheat exposed to Nano-ZnO stress. *Molecules*, 22, 1727. <https://doi.org/10.3390/molecules22101727>

How to cite this article: Xia H, Wang L, Qiao Y, et al. Elucidating the source–sink relationships of zinc biofortification in wheat grains: A review. *Food Energy Secur.* 2020;00:e243. <https://doi.org/10.1002/fes3.243>